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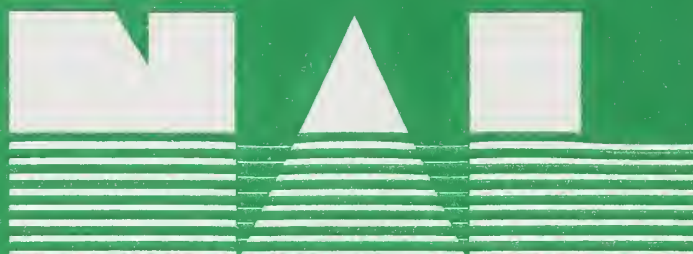
# INFLUENCE OF AGRICULTURAL PRACTICES ON NUTRIENT COMPOSITION OF FOODS

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Proceedings  
Northeastern Agricultural Experiment Station  
Collaborators' Conference  
October 26-27, 1972



**United States  
Department of  
Agriculture**

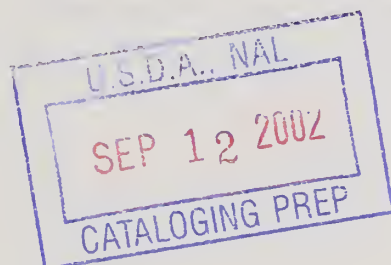


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The Northeastern Agricultural Experiment Station Collaborators' Conference on Influence of Agricultural Practices on Nutrient Composition of Foods, held October 26-27, 1972, at Philadelphia, Pa.. was one of a series of annual collaborators' conferences organized by the regional research laboratories of the Agricultural Research Service, United States Department of Agriculture. The collaborators are staff representatives of the State agricultural experiment stations in each of the four regions. To assure depth and breadth in subject matter, a single area of important research is selected for each conference.

Views expressed in these summaries of papers presented at this conference are not necessarily those of the United States Department of Agriculture. Requests for further information must be sent to the speakers. Mention of commercial products or firms does not imply USDA recommendation or endorsement over others not mentioned.

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## WELCOMING REMARKS

A Summary of a Talk by

Steven C. King  
Deputy Administrator, Northeastern Region  
Agricultural Research Service, USDA  
Beltsville, Md. 20705

Dr. King welcomed the conferees on behalf of the administrative organization of the Northeastern Region of the Agricultural Research Service. He outlined the ARS reorganization put into effect in July 1972. The new Northeastern, Southern, North Central, and Western Regions of ARS coincide with the State Agricultural Experiment Station regions.

A prominent feature of the ARS realignment is that there is a direct, short line of communication from the Administrator through four Deputy Administrators (each in charge of a Region) and on to the Area Directors. A National Program Staff (NPS) of 45-50 persons oriented along lines of commodities and resources will provide needed staff assistance to the Administrator and advice to the Regional Deputies. The NPS support will enable the Deputy Administrators' offices to integrate regional operations with the many needs of ARS as a whole, with other agencies of USDA, and with the States.

Another feature of the ARS reorganization entails the establishment of Regional Planning Committees having equal representation between the States and the Department. Membership on the Regional Planning Committees will include persons from USDA agencies, 1890 Land Grant Institutions, forestry schools, industry, and other groups as the situation makes advisable. Emphasis will be placed on long-range program planning with respect to regional needs.

Dr. King expressed pleasure in being associated with the five research areas of the Northeastern Region and the Experiment Stations. He looks forward to future conferences at Wyndmoor; Beltsville and Hyattsville, Md.; and at Ithaca and Orient Point, N. Y. He is convinced that the Stations, industry and USDA will have a closer working relationship under the new organization than before.



## WELCOMING REMARKS

I. A. Wolff  
Director, Eastern Regional Research Laboratory  
Agricultural Research Service, USDA  
Philadelphia, Pa. 19118

I usually open these annual Collaborators' Conferences with some remarks about the nature and distribution of our program efforts here at the Eastern Laboratory and about the composition and accomplishments of our staff. While I have such information, I'd prefer to discuss it individually with any of you who are interested. Then I can use my short time up here to share with you some thoughts that pertain to the Collaborators' Conferences. First, I'll mention some general aspects of Federal-State cooperation and then some more specific points relative to these conferences.

On March 16, 1972, President Nixon directed to the Congress the first presidential message devoted entirely to the subject of science and technology. I consider this document an interesting and significant indication of public policy. It is recommended reading for any of you who may not have had a chance to see it. One of the major sections of this message is titled "Stronger Federal, State and Local Partnerships." Discussions are proposed in order to "lay the basis for developing a better means for collaboration and consultations on scientific and technological questions in the future." A goal would be to make "government more responsive to public needs" by having each level of government "....do what it can do best."

Dr. Ned D. Bayley, Director of Science and Education in the Office of the Secretary of Agriculture, echoed a similar theme in recent remarks to a group of ARS leaders. Dr. Bayley decreed for ARS an increased ability to relate to the total cooperative program between Federal and State agricultural research groups. Tangible aspects of this thinking may be found in the recent ARS reorganization along regional lines. It is no coincidence that these regions have the same organizational boundaries as the regional groupings of the State Agricultural Experiment Stations. Other specific evidence of current Federal-State interactions lies in the formation and activities of Regional Planning Committees, with which a number of you may already be familiar.

Perhaps we can rightly claim to have some headstart in this matter of Federal-State interactions. Director Wells of the Eastern Regional Laboratory first met with the Eastern Experiment Station Relations Committee in 1940,



and at that time a Memorandum of Understanding was consummated "to provide means for regular consultation and discussion" between the Eastern Laboratory and the Eastern State Experiment Stations on matters of "mutual interest and concern." Ever since, there have been periodic meetings between members of our staff and the Experiment Station Directors or their designees. In 1945, the arrangement was made to have one collaborator from each station visit the Eastern Laboratory once a year. Beginning in 1948 these visits were established as group meetings. This Collaborators' Conference hearkens back to those beginnings.

Perhaps because we have had this arrangement for many years we should scrutinize and examine our procedures to determine if the Collaborators' Conference is meeting the needs we want it to, perhaps to further define those needs, and to make any useful changes that may be indicated.

I presume that most of you know how our topics have been selected in recent years. In a meeting with the AES directors of the region in the spring or summer, we propose several topics that appear to us to be timely and important. This year we proposed "Food Additives - Where are we Going?" "Bridging the Gap between Research and the Application of Research Results," "Physical and Chemical Methodology--Update," and the topic chosen by the Directors - the one we are using today.

I also proposed that the Stations have a greater input into developing the program than they have had in recent years. This year your program committee consisted of the following:

Dr. R. H. Treadway, Assistant Director, ERRL  
Dr. J. Naghski, Chief, Meat, Hides and Leather Laboratory, ERRL  
Dr. J. W. White, Jr., Chief, Plant Products Laboratory, ERRL  
Dr. C. E. Hess, Director, New Jersey Agricultural Experiment Station  
Dr. W. C. Kennard, Associate Director, Agricultural Experiment  
Station, University of Connecticut  
Dr. R. G. Warner, Assistant Director, Agricultural Experiment  
Station and Professor of Animal Nutrition, Cornell University

I want to acknowledge and thank that group for getting together the excellent program we will be enjoying for these two days.

EFFECTS OF CULTURAL PRACTICES  
ON THE COMPOSITION OF FOOD CROPS

F. R. Senti  
Assistant Administrator  
Agricultural Research Service, USDA  
Washington, D. C. 20250

I am happy to participate in this conference which is to cover broadly the influence of agricultural practices on the nutrient composition of our foods. There has been an increased interest in the nutrient composition of foods over the past five years. Among the bench marks in that interest was publication of the result of our Nationwide household consumption food surveys conducted in 1965 and 1966. We analyzed the results of those surveys and arrived at our preliminary analysis a year later, in early 1967. We noted that the composition of the diet, that is the intake of nutrients of people across the Nation, in 1965-66 was poorer, on the average, than it had been 10 years earlier when we made a similar survey

That caused some concern, because we had a more affluent population, yet the foods they were choosing and eating appeared to supply them with less than their needed nutrients. In December 1969, this concern over the nutritional status of our people was reflected in a White House Conference on Food, Nutrition and Health, which raised several questions and made several recommendations about our food, and particularly the feeding of our poor.

Most recently, this concern over nutrition and nutrients in our food has given impetus to requirements for the nutrition labeling of food products. Labeling foods according to their nutrient composition poses a real problem which the Food and Drug Administration is now carefully considering. Bearing directly on this problem is the influence of agricultural practices on the nutrient composition of foods.

There have been many changes in agricultural practices in the past 30 years, particularly since World War II. The public is aware of the increasing use of commercial fertilizers, pesticides, and herbicides, and I think there is a nagging suspicion in many minds that the nutrient composition of our food has changed, and indeed has deteriorated. We do know that our plant breeding programs have been directed primarily to the improvement of agronomic characteristics such as yield, disease resistance, insect resistance, growth habit of the plant, and also (in the case of fruits and vegetables)



to the improvement of such properties as flavor, color, and texture.

This is not to say that attention has not been paid to chemical composition, but I think we would agree that it has been considerably less than the attention given to these other characteristics which are of economic significance. Primary concern for the economic value of food crops in the market is in large measure responsible for this research emphasis.

There are many factors that are known to influence the level of a nutrient in the leaves of a plant, the fruit, or the seeds. These include the soil in which it grows, the season, the particular location, the maturity at harvest, the amount and type of fertilization, and the use of pesticides and herbicides. Location and season include such environmental factors as rainfall, temperature, light intensity, and length of growing season. Because fertilizer application can be controlled and has a marked effect on crop yield, perhaps most attention has been given to the influence of this factor on the chemical composition of plants, including our food crops.

The purpose of this discussion is to point out some of the effects of these factors on yield and nutrient composition of selected food crops. What we really need to know is, How do the conditions of growing crops commercially affect the composition of our foods and thus what impact do they have on the nutrition of our people? This is a hard thing to ascertain because we simply don't have comprehensive data. We can look at controlled experiments where we can see the effects of some of these factors, and in a few cases we can look at the commercial crops, with perhaps some significant data. As one goes through the literature one is impressed with the difficulty of telling just what has happened to the composition of the commercial crop and the food as people eat it, or even as it is supplied to the food processor. I will take a few important food crops and comment on each of them in turn, starting with corn.

## Corn

Although corn is grown primarily as a feed crop, about 120 million bushels are dry milled each year for food use. It is now the most heavily fertilized cereal grain and average U.S. yield has increased 2-1/2-fold since 1947, when statistics on fertilizer use became available. Contributing to the higher yields have been new high-yielding varieties, cultural practices such as high plant populations, and weed control by herbicide application. The question may be asked whether this increase in yield and introduction of new varieties have changed the average protein content of corn.

Influence of nitrogen fertilization and planting rate on yield and protein content of the grain were investigated by Earley and DeTurk (2) in 1948. Table 1 shows the yield increases they found as plants/acre were increased at several rates of nitrogen fertilization. Increased nitrogen had considerably greater effect on yield at higher plant populations. It will

be noted that the yield range in table 1 is from 60 to 184 bushels/acre or a three-fold increase from lowest to highest. Effect on seed protein as given in table 2 was that at a given fertilizer level, protein content decreased with increased plant population and increased yield per acre. However, at each population density, increased nitrogen application increased the protein content of the grain and restored it to that found at lower planting rates.

TABLE 1.--Yield of No. 2 corn as influenced by rate of planting and soil nitrogen<sup>1/</sup>

Planting rate (plants/acre)	Yield (bu./acre at 15.5 percent moisture)			
	N-P <sub>2</sub> O <sub>5</sub> -K <sub>2</sub> O ratio of fertilizer (600 lb./acre):			
	0-14-7	40-14-7	80-14-7	160-14-7
3,920	60	62	69	67
7,841	96	101	109	104
11,682	113	125	135	139
15,682	108	138	149	156
23,522	117	143	155	184

<sup>1/</sup>Earley and DeTurk (2).

TABLE 2.--Percent protein of corn grain as influenced by rate of planting and soil nitrogen<sup>1/</sup>

Planting rate (plants/acre)	Percent protein (nitrogen x 6.25 on moisture-free basis)			
	N-P <sub>2</sub> O <sub>5</sub> -K <sub>2</sub> O ratio of fertilizer (600 lb./acre):			
	0-14-7	40-14-7	80-14-7	160-14-7
3,920	9.9	10.7	10.8	10.7
7,841	9.1	10.0	10.6	10.7
11,682	8.4	8.9	9.8	10.3
15,682	7.8	8.3	9.2	10.0
23,522	8.0	8.0	8.5	9.5

<sup>1/</sup>Earley and DeTurk (2).

Earley and DeTurk concluded that increased planting rate was an important factor in decreasing protein content only when the soil nitrogen was inadequate for maximum yield of grain.

The experiments of Earley and DeTurk were conducted with a single variety



whose protein content appears to be near the average of that for commercial varieties. Data on variation in protein content among varieties are limited, but some indication is given by differences reported for varieties grown in the Illinois yield tests (12). Data for three years indicated that commercial hybrids grown at the same location differed as much as 1.5 percent in protein. These differences were reported not to vary with year; a variety high in protein analyzed high every year while a low protein variety was consistently low.

Varieties of corn have been developed which are relatively high in oil-- 6.8 percent oil, as compared to present commercial varieties which contain about 4.5 percent oil. The relatively high proportion of germ protein in these varieties contributes to higher protein content and to better quality protein of the whole grain. These varieties, however, are not yet widely grown. An important recent discovery has been that a mutant strain designated Opaque-2 contains protein in which the zein component is absent and which is much higher in nutritional quality than common corn varieties. Varieties containing the Opaque-2 mutant gene are not equal in yield to common varieties and are not yet in commercial production in the United States.

Data on the composition of corn received at commercial markets over the past two decades provide a measure of the effect of changes in cultural practices. These data may be compared with average corn yield and rate of fertilizer application. Protein content of corn received by two large corn-processing plants in Illinois from 1947 to 1969 are plotted in figure 1, based on F. R. Earle's unpublished data. Also shown is the average yield for corn in the Corn Belt States which about doubled from 1947 to 1969. Figure 2 relates corn yield to the average rate of fertilizer application for Corn Belt States as pounds/acre of nitrogen on that part of crop that was fertilized. In 1947, 42 percent of corn acres were fertilized at an average rate of 3 lb./acre, whereas 95 percent were fertilized in 1970 at a rate of 112 lb./acre. Protein content changed little over the period. Average for 1947-1951 was 9.66 percent; for 1965-1969 it was 9.80 percent.

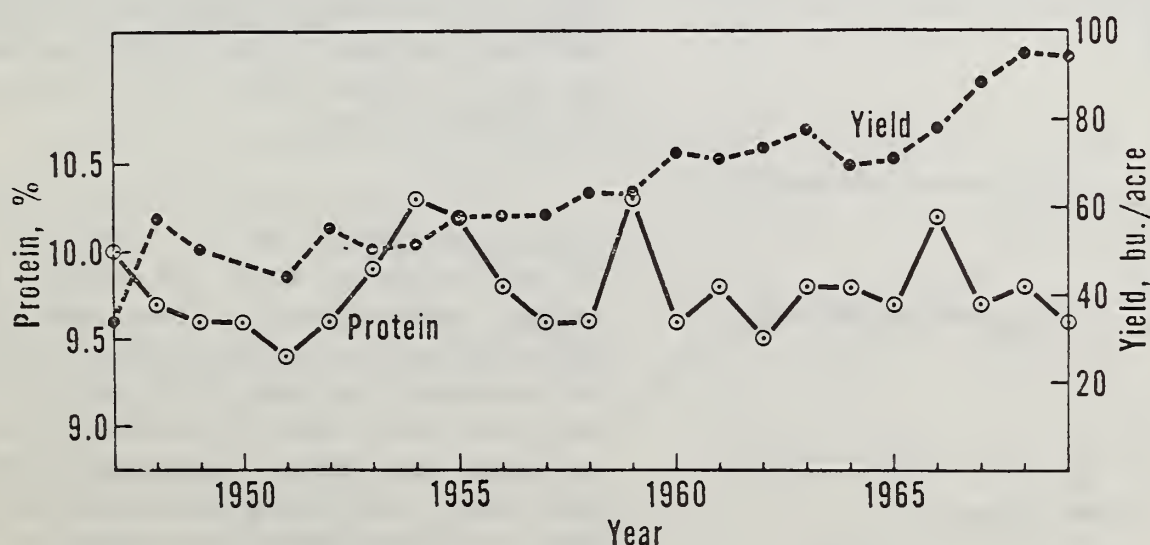


Figure 1.--Relationship of protein content of corn to yield by year, 1947-1969.

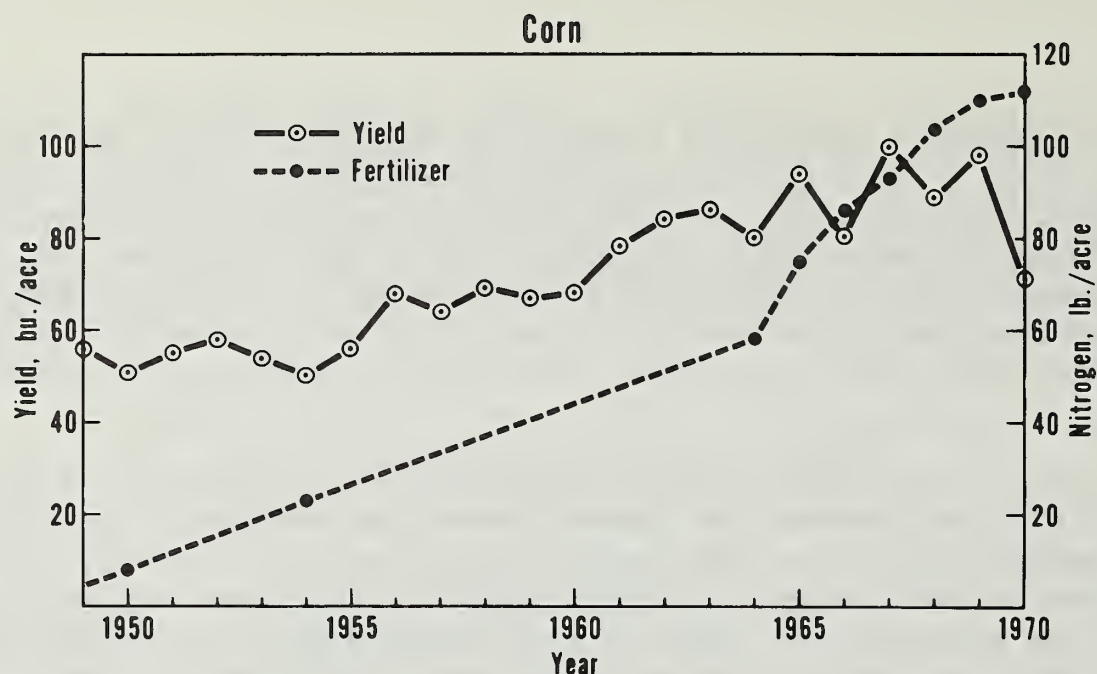


Figure 2.--Relationship of corn yield to rate of nitrogen application in Corn Belt States, 1947-1969.

From the viewpoint of nutritional value, increase in protein content is generally an improvement. However, for present commercial varieties of corn, the nutritional quality of the protein decreases as protein level increases due to increase in the zein component which is relatively low in the essential amino acids lysine and tryptophane, which are limiting in corn. Figure 3 illustrates the relationship between protein level and content of three amino acids including the essential amino acid lysine (13).

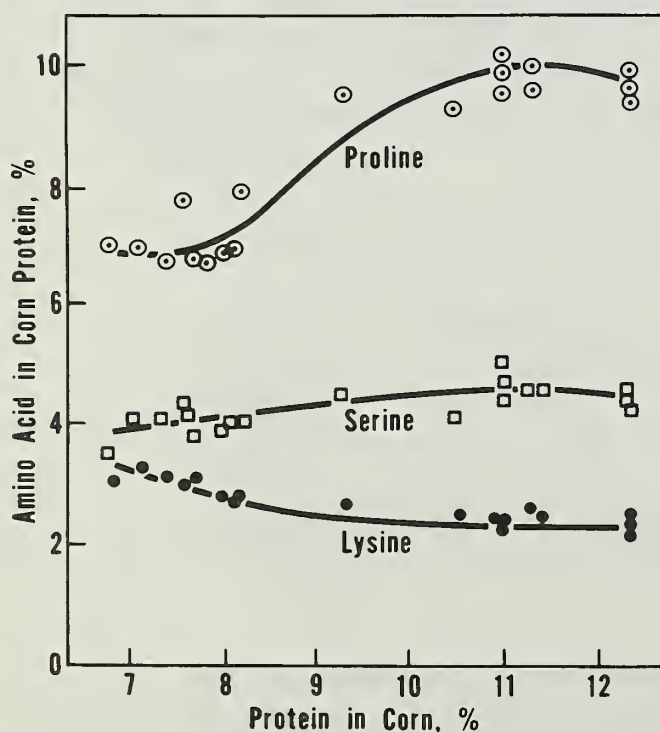


Figure 3.--Variation of amino acid composition with protein content of corn. Sauberlich et al. (13).

## Wheat

Wheat is this country's most important food grain. Although consumption has declined considerably over the past several decades, per-capita consumption is still higher than that of any other single food crop.

As in the case of corn, wheat has changed markedly in the past two decades in two respects that can affect its nutrient composition. These are the varieties grown and rate of fertilization. In contrast to corn, for which breeding programs have been directed primarily to yield and other agronomic properties, new wheat varieties have been selected for the functional properties of their flours. Since protein level and properties are important to functional



properties, we might expect that protein levels of wheat would show no large changes.

Fertilizers, however, which are applied to increase yield can also change the protein content of the grain. This is illustrated by table 3 which shows effects of fertilizer on wheat yield and protein in Kansas (15). Application

TABLE 3.--Fertilizer effects on wheat yields and protein in Kansas, 1948-1952<sup>1/</sup>

N-P <sub>2</sub> O <sub>5</sub> -K <sub>2</sub> O (lb./acre)	Yield (bu./acre)	Protein (percent)
0-0-0	23	12.2
0-50-0	27	11.8
50-50-25 (fall application)	36	12.1
50-50-25 (spring application)	35	12.6
100-50-25 (fall application)	37	13.1

<sup>1/</sup>Smith, F. W. (15)

of 50 pounds of nitrogen per acre applied to winter wheat at seeding in the fall, along with phosphorus and potassium, did not affect protein in the grain. Applied in the spring, however, the same level of nitrogen fertilization increased protein slightly. Yield increase was about the same for fall or spring fertilizer application. Increasing nitrogen fertilization to 100 lb./acre further increased yield only slightly on the average for the 5-year period, but increased protein level about 1 percent over the unfertilized control.

The Kansas State Board of Agriculture has reported protein content of Kansas wheat since 1949. Yield and rate of fertilizer application, the latter since 1954, also are available for Kansas.

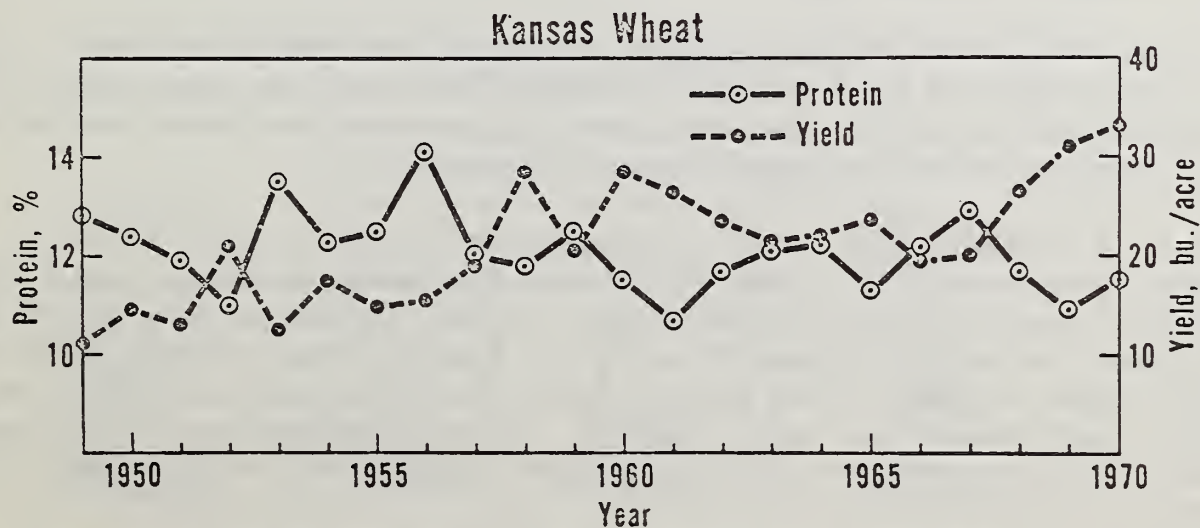


Figure 4.--Average yield and protein content for Kansas wheat, 1949-1970.

Figure 4 shows that average yield of wheat for the State of Kansas has about doubled since 1949, increasing from an average of 14.4 bu./acre for 1949-53 to 25.9 bu./acre for the 5-year period 1966-70. For the same periods, average protein contents were 12.3 and 11.8 percent, respectively, or a decrease of 0.5 percent.

Increased rate of fertilization contributed to the increased yield. As shown in figure 5, nitrogen application increased from 16 lb./acre in 1954 to 46.2 lb./acre in 1970. However, only 21 percent of wheat land was fertilized in 1954 and 51 percent in 1970. Based on the data in table 3, no increase in protein level would be expected.

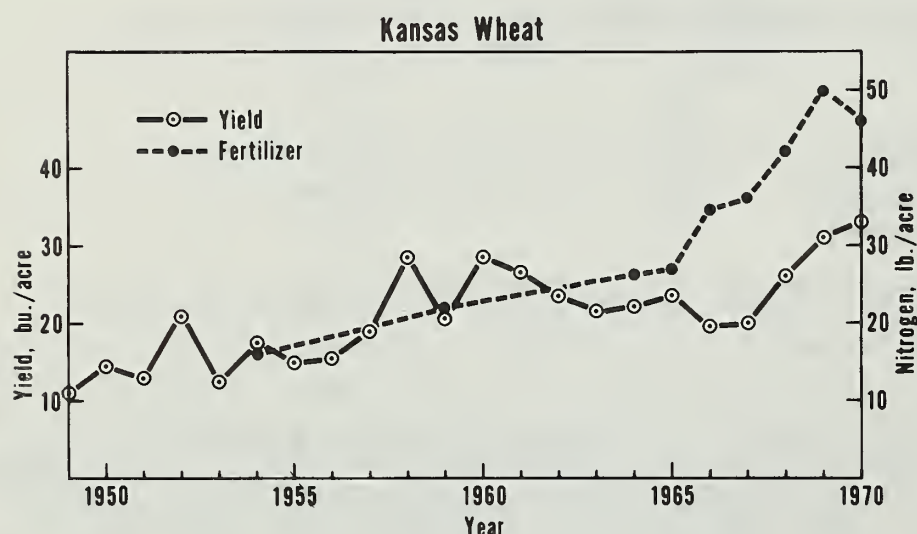


Figure 5.--Yield and rate of nitrogen fertilization for Kansas wheat since 1949.

It should be remembered that the protein values quoted are Statewide averages. Protein content for the same variety varies, however, with location grown. Wheat produced in the eastern half of Kansas generally averages about 1.5 percent lower in protein than that produced in the western half of the State.

Breeding programs (10) for the development of high-protein bread wheat hold promise for raising the protein level one to three percentage points without a lower yield. It would be

desirable from the standpoint of nutritional quality if the lysine content of the protein also could be increased, since lysine is the first limiting amino acid in wheat. To achieve this presents a problem to the breeder, for past experience has shown that lysine content generally decreases with increased protein levels in wheat (18).

So far, only the protein content of wheat has been considered. Wheat is also a good source of many mineral elements essential in human nutrition. The question may be asked whether fertilization and increased yields in recent years have affected the mineral content of wheat.

Table 4 presents the results of experiments (3) conducted in Ohio on the influence of variety, soil, and fertilizer treatment on wheat yield and on the mineral constituents in wheat grain. Three varieties were grown representing hard red winter (Pawnee), soft red winter (Seneca), and white (Cornell 595) wheat classes. Two soils, one a low-fertility acid soil; the other a fertile, well-limed soil were planted. Heavy applications of nitrogen, phosphorus, and potassium fertilizers were applied both singly and in combination.



TABLE 4.--Effect of variety, soil, and fertilizer on the mineral content of wheat<sup>1/</sup>

Mineral	Variety	Soil	Fertilizer
Potassium	**	*	--
Magnesium	--	*	--
Silicon	*	--	--
Manganese	**	**	--
Iron	--	**	**
Zinc	**	--	--
Aluminum	--	--	--
Calcium	--	**	--
Copper	*	**	--

\*\*Statistically significant at 1 percent level.

\*Statistically significant at 5 percent level.

<sup>1/</sup>Data of El Gindy *et al.* (3).

In table 4, asterisks indicate the significance of the effects of variety, soil, and fertilizer treatment on each of the mineral constituents. Fertilizer affected only the level of iron in wheat. Both variety and soil, however, had significant effects on the concentration of a majority of the mineral constituents.

A recent study made by our Consumer and Food Economics Division (23) relates wheat composition to the location grown; associated with location are such factors as soil, climate, crop variety and agricultural practices of the area. Wheat samples were obtained directly from commercial mills in several of the principal wheat-growing States. The samples represented typical commercial blends used at the particular mill. Table 5 presents analytical results for five selected minerals in hard red winter wheats from three different States or regions. The Montana wheat is somewhat lower in nickel and

TABLE 5.--Micronutrients in hard red winter wheat from different regions

Mineral	Concentration of nutrient (p.p.m.)		
	Kansas	Montana	Oklahoma-Texas
Manganese	33.5	38.2	39.9
Copper	5.3	4.9	5.7
Nickel	0.57	0.36	0.47
Zinc	24.3	21.4	18.0
Chromium	0.41	0.33	0.32

copper, and that from Oklahoma-Texas somewhat lower in zinc than wheat from the other States, but generally the wheats are similar in content of the five essential micronutrients.

Table 6 presents analytical results for soft wheats from Michigan, Washington, and Idaho. These wheats appeared to be slightly lower in manganese, copper, and nickel than the hard red winter wheats and somewhat higher in chromium. The Washington sample was considerably lower in copper, zinc, and chromium, but higher in nickel, than the other wheats. Possibly the low zinc content of the Washington wheat may reflect a soil deficiency in zinc which has been reported for the Columbia Basin (14).

TABLE 6.--Micronutrients in soft wheats from different regions

Mineral	Concentration of nutrient (p.p.m.)		
	Michigan	Washington	Idaho
Manganese	32.9	30.2	36.8
Copper	4.7	3.8	4.7
Nickel	0.24	0.39	0.23
Zinc	22.4	12.2	22.7
Chromium	0.48	0.27	0.53

#### Potatoes

Potatoes are a high-yielding crop that receive heavy applications of fertilizer. Potatoes are also an important food in the American diet, having an annual per capita consumption of 120 pounds. An increased amount of potatoes is now going into processed products, amounting to about 50 percent of all potatoes consumed as food. Thus breeding programs in recent years have been concerned with quality factors needed in processing as well as for the fresh product.

Most of the data in the literature relating to fertilization or other practices with potato quality concerns dry matter content. This factor is important to the processing industry since it relates to product yield and "cooking quality" which includes color, flavor, mealiness, and texture.

It has been demonstrated that increasing fertilization reduces the specific gravity and dry matter content of potatoes. However, often "between-years" effects are much greater than "between-fertilizer" effects (20).



It also appears that as percentage of dry matter is decreased, the percentage of crude protein in the dry matter is increased. This is shown by the data (21) in table 7. As nitrogen fertilization was increased, crude protein increased from 9.5 to 12.9 percent of potato solids or about 30 percent. Since percentage increase in protein was greater than the percentage decrease in solids, crude protein also was increased on a fresh-weight basis.

Table 7 also shows that phosphorus content of potato solids was little affected by increased nitrogen fertilization, whereas potassium content decreased about 8 percent.

TABLE 7.--Effect of nitrogen fertilization  
on potato composition <sup>1/</sup>

Nitrogen applied (percent)	Potato composition <sup>2/</sup>			
	Dry matter (percent)	Percent of dry matter		
		Crude protein	P	K
36	22.4	9.5	0.32	1.98
111	21.6	10.9	.32	2.00
186	21.3	11.5	.30	1.86
336	21.3	12.9	.30	1.82

<sup>1/</sup>Wilcox and Hoff (21).

<sup>2/</sup>Fresh-weight basis.

A nutrient of particular importance in potatoes is ascorbic acid--vitamin C. Although the ascorbic acid level in potatoes is considerably lower than in citrus or tomatoes, because of the relatively high consumption rate, potatoes are an important source of vitamin C in the U.S. diet. It is of interest, therefore, to note the effect of various factors on the level of this nutrient. Table 8 compares the effect of nitrogen fertilization rate, location, and year (20). Fertilization had no significant effect. Location grown had an effect which was related to the solids content of the potatoes, but year had the most marked effect. Other investigators (11) have noted similar variation with location, but observed no correlation with solids content, and less variation with year. Ascorbic acid levels in 5 varieties grown for two years at 5 locations ranged from 11 to 27 mg./100 g. fresh weight, but there were not significant differences among varieties.

While the Northeast continues to be an important growing area for potatoes, the Nation's potato production has increased mainly due to production increases in the Pacific Northwest and the Red River Valley of North Dakota-Minnesota.

TABLE 8.--Effect of nitrogen fertilization, location, and crop year on ascorbic acid content (mg./100 g. fresh wt.) of Norland potato tubers<sup>1/</sup>

Year	Nitrogen rate	Location		
		A	B	C
1960	0	13.1	14.8	17.3
	30	12.1	14.6	17.0
	60	14.1	14.2	16.7
1961	0	25.2	21.7	19.5
	30	26.4	21.4	18.4
	60	26.0	21.0	18.0

<sup>1/</sup>Teich and Menzies (20).

### Citrus

Citrus fruit is a major source of vitamin C in the U.S. diet. Citrus is grown on sandy, infertile soils in Florida that have required fertilization with a number of minerals in addition to the usual N, P, and K to avoid deficiencies and produce high yields. Soils in the West and Southwest used for citrus production have had relatively high fertility compared to Florida citrus-orchard soils. Nitrogen has been the principal fertilizer used except that in California sprays containing zinc are used for the control of zinc deficiency.

In an excellent chapter on citrus nutrition P. F. Smith (16) has described the effects of fertilization with various elements on citrus fruit quality and composition. He reports that the effects of increased nitrogen application on the soluble solids, acids, and vitamin C content of the fruit is of relatively small magnitude--of the order of 3 to 5 percent. Increased phosphorus fertilization affects these quality factors to a slightly greater degree. Potassium fertilization has more effect on fruit quality than nitrogen or phosphorus. Fruit size and acidity of juice are strongly increased by potassium fertilization. Vitamin C content tends to be increased.

Rate of both phosphorus and potassium fertilization have been reduced in Florida as compared to the practice in 1940-1950. Use of heavy applications of phosphorus resulted from early grower experience with benefits from bone meal and fish scraps as phosphorus sources, but it is likely that these also carried trace minerals that were responsible for much of the benefit. Potassium has been reduced from 150-300 lb./acre in the 1940's to the present 100-200 lb./acre.

Table 9 shows how the ascorbic acid content of commercial orange juice



has been affected by changed fertilizer practice and any other practices that may have changed or been introduced in the citrus orchards since 1936-38.

TABLE 9.--Vitamin C content (mg./100 ml.) of Florida orange juice, 1936 to 1971

Harvest	1936-38 <sup>1/</sup>	1956-58 <sup>2/</sup>	1970-71 <sup>3/</sup>
Midseason	40-50	38-50	46-52
Late season	30-42	32-43	--

<sup>1/</sup>Hand-reamed.

<sup>2/</sup>Commercial pack frozen concentrated orange juice.

<sup>3/</sup>Commercial pack chilled orange juice.

Frozen concentrated orange juice had not been commercialized in 1936-38 and data for these two seasons were on hand-reamed orange juice from a sampling of the crop (6). Data for 1956-58 (7) are for commercial pack frozen concentrated orange juice, whereas those for 1970-71 are for commercial pack chilled orange juice. The latter, from M. K. Veldhuis' unpublished data, represent a limited number of samples and may not be as representative of the crop as are for the data for earlier years. From these data we would conclude that there has been no significant change in the ascorbic acid content since the mid-1930's. In contrast to many other crops, there has been little varietal change in juice oranges. The midseason oranges harvested mainly in January and February have been principally the Pineapple orange, while the Valencia orange has been the principal late-season variety harvested in March and April.

As mentioned earlier, successful citrus culture in Florida requires fertilization with trace minerals as well as with N, P, and K. Copper, zinc, manganese and iron are commonly applied, frequently by foliar spray. In view of the known soil deficiencies in these elements, it is of interest to compare mineral analyses (17) of oranges in 1940 with those for 1953 as given in table 10. Phosphorus, potassium, and calcium content of the whole fruit was less in 1953 than in 1940. The decrease in phosphorus and potassium may reflect the change to a lower rate of fertilization with these elements, as mentioned earlier. The level of trace minerals, except copper, was increased. Unfortunately, we have no data on mineral composition for more recent crop years, nor has it been reported whether the change in trace minerals from 1940 to 1953 could be correlated with change in fertilizer application practice.

#### Tomatoes

The factors affecting vitamin C level in tomatoes were the subject of intensive investigations a few years ago (4). Some of the important findings

TABLE 10.--Mineral composition of Valencia oranges<sup>1/</sup>

Mineral	Content (lb./2000 lb. whole fresh oranges)	
	1940	1953
P	0.50	0.41
K	5.09	3.95
Ca	1.40	.74
Mg	.24	.36
Mn	.0017	.0022
Fe	.005	.008
Cu	.0014	.0014
Zn	.0021	.0043

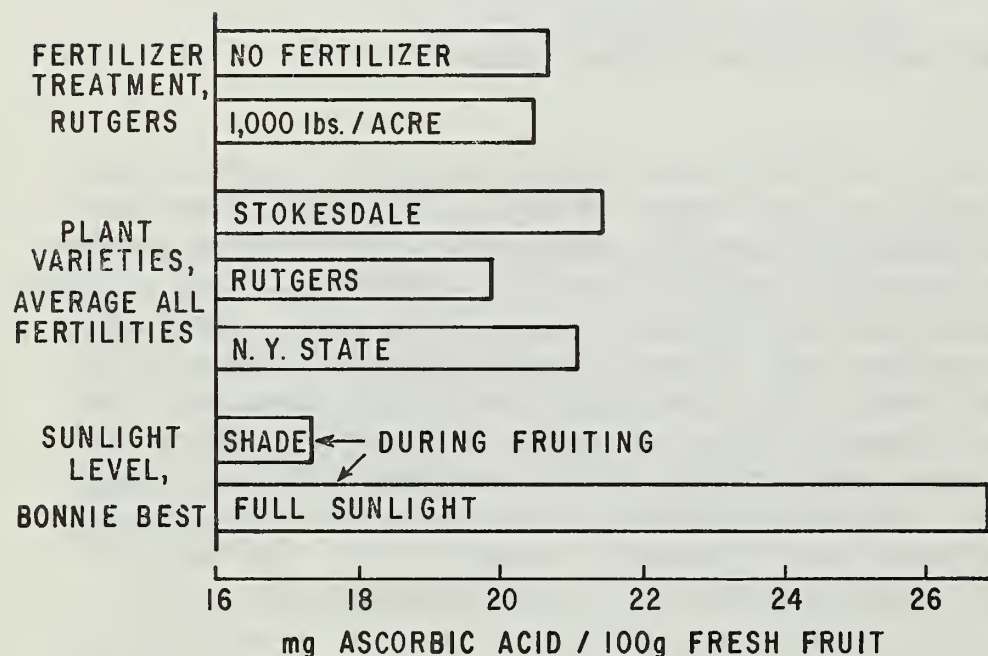
<sup>1/</sup>Smith and Reuther (17).

Figure 6.--Effect of fertilizer application, variety, and sunlight on ascorbic acid content of tomatoes.

are illustrated in figure 6. Fertilizer had little effect on vitamin C content, nor was there much difference among the Stokesdale, Rutgers, and New York State varieties. The amount of sunlight received by the plant, however, had a major effect. Plants of the Bonny Best variety shaded during fruiting had a vitamin C content of 17 mg./100 g. of fruit, whereas fruit of plants grown in full sun contained 27 mg./100 g. The effect appears to depend on the amount of light striking the fruit.

Hamner and Maynard (5) report on work by Vesselkind and others in Russia indicating that tomatoes protected from light by enclosure in black paper sacks contained only about 1/5 the ascorbic acid content per unit weight of fruit produced in light. Brown (1) also noted that tomatoes receiving direct sunlight were higher in ascorbic acid than fruits shaded by leaves or artificial cover.

From other reports, it appears that immature, green tomatoes are lower in vitamin C than ripe ones, but that after a fruit has reached its full size and is in the so-called mature-green stage, the increase in vitamin C during subsequent ripening is slight. The practice of harvesting tomatoes at the mature-green stage, which has been followed to improve shipping properties, thus does not greatly reduce the vitamin C content of the fruit.



Tomato varieties higher in vitamin C than those shown in figure 6 have been released. The variety Doublerich developed at the New Hampshire Agricultural Experiment Station had vitamin C levels above 50 mg./100 g. fresh fruit, as compared to 15-25 mg. for ordinary varieties. Potential of varietal development on nutrient composition is also illustrated by development of the Caro-Red variety at Purdue University, which produces fruit containing approximately ten times the pro-vitamin A content of common red tomatoes. These values are within the lower ranges reported for carrots. Although these two varieties are not now widely grown, they are good sources of germ plasm to satisfy future interest in high-vitamin tomatoes.

## Effect of Nitrogen Fertilization

### on Nitrate and Nitrite

### Content of Vegetables

Plant species differ in their tendency to accumulate nitrate in their tissues. In most plants, nitrate--the major form in which nitrogen enters the root system--is rapidly reduced to ammonium ion, thence synthesized into amino acids and proteins and into other nitrogenous constituents. The process of protein synthesis in some plants may be affected by stresses applied from unusual climatic conditions such as drought or prolonged cloudy weather. Or, the synthesis may be in response to high soil nitrogen levels, perhaps resulting from high rates of application of nitrogen fertilizers. Cereal grains used at an immature stage for pasture or hay, corn at the silage stage, and sudan grass are among the forage crops that have been found to accumulate high nitrate levels and to have been involved in cases of nitrate poisoning of ruminant animals.

Although the condition is referred to as nitrate poisoning, toxicity is due to nitrites. Nitrate itself is relatively nontoxic and it is reported that one gram is a single safe dose for all humans. Under appropriate conditions, bacterial action in plant material after harvesting can convert nitrate to nitrite.

Nitrate can also be converted to nitrite after ingestion by action of microflora in the digestive tract. In human adults this occurs in the lower intestines where the nitrite is not absorbed, but in young infants with certain digestive ailments, reduction may occur in the stomach or duodenum, where the nitrite can be absorbed, causing illness.

Among the vegetable crops, spinach and beets have been reported to be relatively high in nitrate content. Recently, the effect of nitrogen fertilization on the nitrate and nitrite content of beets and spinach (table 11) has been studied (8). For beets, there was a strong increase in nitrate nitrogen at rates of nitrogen fertilization between 100 and 200 lb./acre, with a lesser proportionate increase above and below these levels. Nitrate nitrogen was very low in beets regardless of fertilizer level, having a maximum value of 2 p.p.m.

TABLE 11.--Effect of fertilization on nitrate and nitrite content of table beets and spinach<sup>1/</sup>

Vegetable	Fertilizer (lb. N/acre)	Nitrate-N (mg./100 g.)	Nitrite-N (p.p.m.)
Beets	0	40	0.9
	50	51	1.7
	100	110	1.5
	200	310	1.2
	400	350	2.0
Spinach	0	248	0.5
	400	690	0.6

<sup>1/</sup>Lee et al. (8).

Data were reported for spinach at only two nitrogen fertilization rates--0 and 400 lb./acre. Compared with beets at these fertilization rates, spinach accumulated considerably higher nitrate levels. Nitrite level in spinach was very low and unaffected by fertilizer application.

It is difficult to ascertain the significance of these fertilizer-response data to commercial crops, since farm fertilizer application rates may vary considerably. Lee (9) has reported a comparison of analyses of vegetables collected at the Geneva, New York, market in 1949 and in 1971. These are presented in table 12. The data indicate that nitrate content increased over this period in beets, broccoli, radishes, and spinach received at the Geneva market, but decreased in cabbage, carrots, cauliflower, celery, and lettuce.

TABLE 12.--Nitrate nitrogen in fresh vegetables<sup>1/</sup>

Vegetable	Nitrate N (p.p.m.)	
	1949	1971
Beets	306	548
Broccoli	180	214
	884	
Cabbage	511	207
Carrots	101	76
Cauliflower	460	238
Celery	278	226
Lettuce	287	63
Radishes	70	456
	270 <sup>2/</sup>	
Spinach	70	468
	836 <sup>2/</sup>	

<sup>1/</sup>Lee, C. H. (9). Data for 1949 cited by Lee from article by C. H. Wilson in Agron. Jour. 41, 20 (1949).

<sup>2/</sup>Nitrate content of juice.



## Effect of Pesticides on Plant Composition

Since herbicides have a marked effect on seed germination and plant growth of susceptible species, it may be questioned whether a more subtle, yet significant, effect on nutrient composition of resistant species may result from application of these chemicals. The same question can be posed with respect to pesticides generally.

It has been reported that some fungicides increased the sugar content of apples, but that residues of aldrin and dieldrin in roots of carrots had no effect on the ascorbic acid or sugar content of this vegetable. Simazine, a widely used herbicide, has been reported to increase the protein content of several crops, including beans, peas, sweet corn, rice foliage, rye grass, and alfalfa. Sweeney and Marsh (19) reported that the carotene content of Danvers and Chantanay carrots (table 13) grown in soil treated with the herbicides Linuron and CIPC was significantly increased, as was the carotene content of butternut squash grown in soils treated with Amiben or Dinoseb. On the other hand, soil treated with the herbicide CDEC decreased the beta carotene content of two spinach varieties tested, whereas Endothal increased beta carotene in one variety but decreased it in the other.

TABLE 13.--Effect of herbicides on carotene content of carrots, squash, and spinach<sup>1/</sup>

Herbicide	Carotene content (µg./100 g.)					
	Carrots		Squash		Spinach	
	Danvers <sup>2/</sup>	Chantenay <sup>2/</sup>	Butternut <sup>2/</sup>	Hubbard <sup>3/</sup>	Bounty <sup>3/</sup>	668.9 <sup>3/</sup>
None (control)	7,890	5,020	2,090	820	4,830	3,870
Linuron	9,560	7,390				
CIPC	10,220	6,940				
Amiben			2,620	960		
Dinoseb			2,740	850		
CDEC					4,220	3,610
Endothal					3,910	4,560

<sup>1/</sup>Sweeney and Marsh (19).

<sup>2/</sup>Total carotene.

<sup>3/</sup>β-carotene.

The effect of soil fumigants on the carotene content of carrots also has been investigated. The control of nematodes in fields used for carrot production is presently accomplished by means of fumigation with chemicals which include Telone and Nemagon. In studies covering three crop years, it was found that fumigation with these chemicals increased the carotene content between 20 and 40 percent over the check samples (22).

Increased use of organic pesticides has introduced residues of some of these chemicals, but since these are not nutrients, discussion of these residues is beyond the scope of this paper.

### Conclusion

By and large, the evidence would indicate that changes in agricultural practices over the past 20 to 30 years have had relatively small effect on the average nutrient composition of our food and feed crops. There is indication, for example, that protein content of wheat in certain areas may have decreased slightly; on the other hand, there is evidence that the protein content of corn has increased slightly.

The evidence would indicate also that varietal shifts and fertilization levels may have some effect; yet, these are generally smaller than the variation by year or by location. In view of the effect of location, shifts in growing areas of crops, such as potatoes, that have occurred over the past two or three decades may have introduced some compositional changes, particularly with respect to mineral elements. Data are sparse on this point, however. There is indication that zinc deficiency has developed in soils of certain regions, and this may be reflected in a lower concentration of zinc in plants such as wheat grown in the Columbia River Basin. As to the increased use of pesticides over the last two or three decades, most of the evidence indicates that any effect these chemicals may have on composition is to increase, rather than to decrease, the level of nutrients that have been examined.

It is apparent from this review that more systematic sampling of crops and more extensive analyses for more of the essential nutrients is required to provide needed information on food composition and impact of future changes in agricultural practices.

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## ORGANIC FARMING AND PLANT COMPOSITION

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There seems to be little doubt that a relationship exists between soils and the nutritional quality of food crops. However logical this may appear, the relationship is not simple. It has been recognized for a long time that factors such as climate, location, harvest time, storage handling, and variety and genetic variables are strong determinants in the composition of foods reaching the market (1, 2, 3).

Over the years much work has been reported on the effects soil amendments, particularly inorganic chemical fertilizers, have on yields of crops (10). Much less has been done on nutritional composition. It was generally accepted that large, healthy-looking crops were synonymous with plants of high nutritional value. There is some validity to this belief. In addition to yield response, occasional measurements were made for several nutritional components--carbohydrates, fats, proteins. It was also recognized early that mineral and vitamin content are important indicators of nutritional quality. Assays were normally a combination of chemical analyses and animal feeding trials.

Often in fertilizer yield experiments, animal manures were included for comparison with chemicals. This indicated an early recognition of possible differences between the two sources of mineral elements. Results have been published widely and in the main show that there is little difference in crop response between the two fertilizer management systems. Generally, manures maintained or increased soil organic matter content, improved soil structure, and tended to encourage higher water-holding capacity.

Much less has been reported on the possible effects the two fertilizer sources might have on nutritional quality. It has been almost universally accepted that the use of manures is good practice for improving soil conditions and plant growth for a balanced agriculture, but is not necessarily related to nutritional quality. Traditionally, orthodox agriculture in this country has always encouraged the use of organics wherever possible and practical.

However, during the last decade there has been a very strong and insistent interest in a system of agriculture known as Organic Farming. Advocates



of this concept avoid the use of any manufactured chemical fertilizers, pesticides or food additives in the growing and marketing of crops. Limestone, sulfur, ground granite rock, rock phosphate, etc., are accepted soil amendments. A basic tenet of organic farmers is that foods grown using organic manures or composts, and other nonchemical controls such as companion cropping and natural pesticides as rotenone and pyrethrums, produce plants of greater nutritional and better health-giving values than those grown with chemicals.

A very important consideration in the organic way is a deep concern and desire for a simpler, less stressful, and more basic life style in close harmony with a natural, chemical-free environment. One very useful byproduct of this movement is a commitment to the reduction of waste and an effort to recycle as much organic matter as possible. Much is made of specially and carefully prepared composts using almost all degradable organic materials wherever available. Allied with this holistic philosophy are special kinds of foods, cooking techniques, and diets. However, the social life-style aspects of organic farming are beyond the scope of this discussion, although there are attitudes and values involved which probably should not be separated from the process of growing plants by the organic farming system.

There are many unsupported and fragmentary articles in popular-type magazines and brochures which claim that foods produced by this system differ in nutritional composition. Very few results have been published in generally-accepted scientific journals on the effects organic farming methods may have on nutritional quality. There are a number of reports on the use of manures, but these results, in actuality, represent comparisons of systems other than what is commonly called organic farming. Certainly there is some relationship, but care must be exercised in drawing comparisons from such results.

As examples, early reports from Europe show no consistent differences in the accumulation of phosphorus and potassium in potatoes from additions of manures and chemical fertilizers. Nitrogen, however, did increase where chemicals had been used. Leong in 1939 showed the highest vitamin B<sub>1</sub> content in barley where manure alone had been applied, but a higher vitamin B<sub>1</sub> content in wheat where the manure was supplemented with minerals (7). Heavy applications of manure increased vitamin B<sub>1</sub> in some varieties of wheat in England (4). Swanson in Iowa, comparing the effects of chemicals and manure in sand culture, showed little difference in vitamin A in sweetpotatoes (12). Ijdo, in several years of experiments in Holland, showed inconsistent differences in vitamins A and B<sub>1</sub> in spinach resulting from organic sources of nitrogen (6). In some years, spinach in check plots (no fertilizer) had equally high vitamin content. Some brief observations in India show increases in vitamin C in spinach associated with farmyard manure (9). In results listed above, experiments were of a rather limited scope and do not lend themselves to critical interpretation.

In 1950, Brandt and Beeson at the Nutritional Laboratories in Ithaca, N. Y., reported that manure treatments did not significantly favor accumulations of ascorbic acid and carotene in rye or potatoes, nor did they produce any significant changes in iron and copper content (5). A few comparisons

were also made using composts. In both carrots and green snap beans the composts made little difference in the ascorbic acid and carotene content. Carrots grown in a different type of soil and treated with chemicals in the same trials accumulated significantly more carotene than those treated with the compost. The difference was attributed to the larger size of the carrots related to the soil type.

At about the same time Schuphan in Germany compared the carotene content of tomatoes and carrots grown using manure alone and manure supplemented with minerals (11). There is some indication that the mineral additions increased vitamin content. Also, infants fed vegetables from the fortified manure plots showed an increase in vitamin A in blood serum and about a 20-percent increase in weight gain per day.

In a newsletter published in 1972, Arrowhead Mills, Hereford, Texas, marketers of whole grain flour under the Deaf Smith Organic Farms label, published results of chemical analyses on wheat from seven farms classified as "organic." When results are compared with average figures in the USDA Handbook on Composition of Foods (13), little difference can be noted. Comparisons were made for a number of nutritional components including proteins, phosphorus, calcium, zinc, and other metallic elements.

There are a few reports from private laboratories on the special handling of organics in agricultural systems. Closely related to the organic farming movement is a group known as Biodynamic Farmers. This had its philosophical origin notably from Dr. Rudolf Steiner in Germany and was established in this country under the leadership of Dr. E. Pfeiffer with laboratories and experimental farms in Spring Valley, N. Y. (8). The biodynamic concept and philosophy are much more concerned with specially prepared composts and viable, profitable farming than they are with gardening, with which organic farming is often confused. Pfeiffer's data strongly suggests that biodynamic composts produce nutritionally better plants than conventional chemical fertilizers. His experiments are scanty and have not been repeated or tested by other laboratories, but some are rather ingenious and unusual and should not be brushed aside without question.

The ideas expounded by organic farmers are important and raise serious questions. To adequately test their values it is suggested that recognized agricultural laboratories and stations set aside some of their research capabilities to check more carefully the validity of some of the claims.

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EFFECTS OF STORAGE, TRANSPORTATION, AND MARKETING  
CONDITIONS ON THE COMPOSITION AND NUTRITIONAL  
VALUES OF FRESH FRUIT AND VEGETABLES

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Fruits and vegetables are important sources of a number of vitamins and minerals in our diet. They may also supply significant quantities of the major components such as carbohydrates and proteins. The changes and losses in some of these constituents in fresh fruits and vegetables after harvest may be important if the movement of the commodity from the producer through the transport, storage, and marketing chain to the consumer is unduly long or if conditions during this period are unfavorable for the commodity.

Fresh fruits and vegetables are living tissues when harvested and retain vital functions until senescence and decay, or processing, stop them. Enzyme activity plays a major role in any changes in composition in these tissues and is greatly influenced by the temperature and humidity conditions to which the fruits and vegetables are subjected. Thus temperature and humidity are two big factors to be considered in any after-harvest handling of fruits and vegetables. Obviously any handling implies careful avoidance of mechanical damage.

The period of transport may be regarded as a portion of the storage period. The wholesaling, retailing, and the holding periods in consumers' homes are variable and difficult to standardize in experimental tests. Some storage studies have included a final holding stage at conditions differing from those of the main storage period in order to simulate the effects of the last steps in the marketing chain.

Many fruits and vegetables ripen during storage. Since ripening is a general phenomenon influenced primarily in its rate by storage conditions, no detailed discussion of the many changes in composition associated with ripening is included here.

Storage may affect the vitamin content, some of the major constituents such as carbohydrates, and the conversion of some compounds to toxic forms.

Newer storage techniques such as the use of controlled atmospheres may have an influence on the composition of fresh fruits and vegetables. A discussion of these topics will form the remainder of this presentation.

### Ascorbic Acid

Vitamin C or ascorbic acid is one of the more important nutrients supplied by fresh fruits and vegetables and is also one of the most sensitive to destruction when the commodity is subjected to adverse handling and storage conditions. Unfavorable conditions involving high or low (chilling) temperatures, low humidity or wilting conditions, and physical damage produce stress conditions which accelerate the oxidation and loss of ascorbic acid.

The loss of ascorbic acid is associated with the degree of wilting in kale (4). The same degree of wilting causes much higher losses at 50° and 70° F. than at 32°. Other leafy vegetables such as spinach, turnip greens, and collards respond similarly. Cabbage, which is also a leafy vegetable, loses ascorbic acid more slowly than some of the previously mentioned vegetables. Whether this is because of its structure or because of a protective mechanism is not clear.

The loss of ascorbic acid in peas and beans may be retarded by storing these vegetables in the pods. Shelled lima beans lose ascorbic acid at twice the rate of unshelled beans at the same temperature (2).

Potatoes, although relatively low in ascorbic acid content, have been considered a rather important source of vitamin C in the diet because of their widespread consumption. This significance has undoubtedly changed with the increase in the more highly processed forms of potatoes such as chips and french fries. Many investigators have found that ascorbic acid content of fresh potatoes decreases during storage and that the loss is greater as the storage temperature is reduced below 50° F. Potatoes stored at 40° have been found to lose as much ascorbic acid in two months as comparable lots stored at 50° or 60° lost in five months.

Many fruits, particularly citrus, are good sources of vitamin C. There are conflicting reports on the stability of ascorbic acid in citrus. It is generally agreed that lemons retain nearly 100 percent of their ascorbic acid during storage and that grapefruit also lose very little ascorbic acid. Reports differ for oranges and tangerines, but it has been concluded that loss of vitamin C in fresh citrus is unlikely to exceed 10 percent under reasonable conditions of distribution and marketing (8). Many fruits and fruit products are used in the fresh or fresh frozen form, thus most of their ascorbic acid content is directly available and not partially destroyed in the cooking process as is more customary for vegetables.



## Folic Acid

Folic acid is also lost to a considerable extent if storage conditions are unfavorable. The legume seeds, asparagus, spinach, turnip greens, and some other leafy vegetables supply significant quantities of folic acid to the diet. These vegetables stored for two weeks at refrigerator temperatures or, still better, in crushed ice were found to lose little or none of their folic acid content. However, storage at room temperature for three days resulted in losses of 50 to 70 percent of the vitamin (6).

## Thiamine and Riboflavin

Fruits and vegetables are not considered important sources of thiamine and riboflavin but some of the legume seeds may supply significant amounts of thiamine. Fresh green shelled lima beans stored at room temperatures for two to four days were found to retain 90 percent of their thiamine and nearly an equal percentage of riboflavin (2). Thus, these vitamins were found to be relatively stable under unfavorable commodity storage conditions.

## Carotene

It has been shown that carotene content of some vegetables such as sweet-potatoes may actually increase during storage (3). On the other hand, leafy vegetables subjected to wilting conditions may lose one-half to two-thirds of their carotene when held at or near room temperature for four days (5).

## Carbohydrates

The changes in sugar content in freshly harvested sweet corn is often referred to as a classic example of the effect of temperature on changes in composition of a freshly harvested commodity. Recent surveys reported from Pullman, Wash., on the commercial handling of sweet corn for processing show that transit and holding times of the loads ranged from 3-1/2 to 20 hours, the average temperature of the loads from 50° to 90° F., and the loss in sugar from 1 to 27 percent (11).

The hydrolysis of a portion of the starch to sugar in potato tubers stored at temperatures below a certain level, usually about 50° F., is another example of the effect of temperature on composition.

Many other changes such as the synthesis and hydrolysis of proteins and changes in pectic constituents during storage could also be described, but it is doubtful that these changes have any real nutritional significance. Some of the changes may have marked effects on the acceptance quality. The loss of sugar in sweet corn and its increase in potatoes are both detrimental to

quality, while the increase in sugar in sweetpotatoes and parsnips is considered beneficial.

### Nitrates and Nitrites

Another change that may occur during storage of some commodities is a conversion of nitrates to nitrites. Recent studies here at our host laboratory and at other locations emphasize the seriousness of these changes. It has been shown that heavy application of nitrogen fertilizer will increase the nitrate concentration in spinach and that 10 to 15 percent of the nitrates may be converted into nitrites after spinach has been held at room temperature for four days (10). Other studies with beets and spinach (9) and potatoes (7) have shown that the conversion to nitrites is minimal or non-existent in beets and spinach when they are stored at 35° F. or when potatoes are in normal storage. However, potatoes held under low oxygen concentrations showed a slight increase in nitrites.

### New Storage Techniques

So we may ask, What new storage techniques are being used today that have an influence on the composition or nutritional quality of fruits and vegetables? The use of modified or controlled atmospheres for prolonging the storage of apples has been a commercial practice for a number of years and has been expanding for apples and for other commodities in recent years. A study in France (1) indicates that apples stored in 3 percent oxygen and 97 percent nitrogen at 15° C. retained over 60 percent of the original ascorbic acid after three months of storage, whereas those stored in air retained less than 20 percent. When similar tests were conducted at a more normal storage temperature of 4° C., no differences in ascorbic acid retention were noticeable. Atmospheres modified by the addition or the controlled accumulation of carbon dioxide have been found to aid in retaining the sugar content of sweet corn and the tenderness of asparagus.

The use of hypobaric or low-pressure storage for fruits and vegetables is being investigated in several laboratories. Its effectiveness in prolonging the storage life of some commodities is evident, but the influence on composition is not known at this time.

The losses in nutritive value of fresh fruits and vegetables during storage, transportation, and marketing are not likely to be great if the following are observed:

1. Care is taken to keep physical damage at a minimum during handling.
2. The time between harvesting and receipt by the consumer is not too long.



3. The temperature and humidity conditions are kept near optimum for the commodity.

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## GENETIC ENGINEERING TO REMOVE UNDESIRABLE COMPOUNDS AND UNATTRACTIVE CHARACTERISTICS

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Traditionally the greatest emphasis in plant breeding has been placed on increasing our total food supply. As a result of the success of this plant breeding research, food shortages are considered less of a threat throughout the world today than in the 1960's. Consequently, the average well-fed American today would probably give top priority to the problems of consumer acceptability and food safety.

### Naturally Occurring Toxicants in Plants

The recent public health concern has arisen from the growing realization that many foods contain naturally occurring toxic or deleterious properties. As new germ plasm is used to breed vegetables and other food crops with increased resistance to insects and diseases, there is real danger of increasing the levels of these undesirable substances. For example, a potato variety released a few years ago because of its superior processing qualities had an unusually high content of total glycoalkaloids. Likewise, some potato cultivars are insect resistant because they contain a high content of a cholinesterase inhibitor.

Although there is great potential public health significance to the presence of substances such as alkaloids, antimetabolites, oxalates, and allergens in foods, not nearly enough research has been devoted to the genetic or physiological factors that determine their synthesis and ultimate level in plants. Because research in these areas is relatively new, the sum total of genetic knowledge developed and recorded in the literature is frustratingly sparse. Few reports are to be found that define an association between genotype and undesirable or unattractive characters. We might note that most of the papers on this subject have been published since 1970.

## Nitrates

The hazard of excessive nitrates in foods has been generally recognized, especially for infants (33).

Nitrates can be present in significant quantities in some nitrate-accumulating vegetables grown in nitrogen-rich soils and in circumstances such as reduced light and water. Under these conditions, lettuce, turnips, greens, collards, dryland cress, beets, radishes, celery, endive, kale, and parsley can have relatively high concentration of nitrates (16). On the other hand, the concentration in tomatoes, potatoes, carrots, beans, peppers, sweet corn, and peas is quite low, even in nitrogen-rich soils. Viets and Hageman in making a study in 1971 of factors affecting the accumulation of nitrates found little or no specific data in the literature concerned with differences in nitrate absorption as a function of genotype (31). However, genetic control of nitrate content in vegetables has recently been demonstrated. Small but consistent differences were found in nitrate content of the spinach cultivars Winter Bloomsdale, Northland, and Tuftguard fertilized at four rates of nitrogen (3).

Winter Bloomsdale, a savoy-leaved cultivar, accumulated nearly twice the nitrate found in Northland and Tuftguard, the two smooth-leaved cultivars, indicating that the uptake of nitrogen may be accidentally associated with the genes for savoy-leaf. Savoy-leaf is partly dominant over smooth leaves.

The association of nitrate N uptake and leaf character was further verified by Maynard and Barker in 1972 (20) as follows:

TABLE 1.--Association of nitrate N concentration with leaf type in spinach

Leaf type	No. of cultivars	Nitrate N (percent dry wt.) <sup>1/</sup>	
		Petioles	Blades
Savoy	6	1.14 <sup>b</sup>	.30 <sup>b</sup>
Semi-savoy	6	.90 <sup>b</sup>	.18 <sup>ab</sup>
Smooth	6	.42 <sup>a</sup>	.08 <sup>a</sup>

<sup>1/</sup>Data in a column followed by same letter are not significantly different at the 5 percent level of probability.

However, Cantliffe (5) could not detect consistent differences in nitrate between savoy- and smooth-leaved cultivars, although he did find that under conditions of nitrate fertilization the cultivar Virginia Savoy contained more nitrate than the smooth-leaved cultivar Northland. He concluded that the



variation in nitrate content was not caused by differences in nitrogen uptake because both cultivars contained a similar concentration of total nitrogen. The differences in nitrate content could have been caused by a genetic difference in nitrate reductase activity between the two cultivars, with no real association between leaf type and nitrate reductase activity.

In a cooperative study involving seven universities, cultivar characteristics were a major factor controlling nitrate accumulation in three vegetables (11).

Nitrate content in plants varies inversely with the level of nitrate reductase activity in the same tissue (15). The level of this reductase activity is highly heritable (27). These findings demonstrate the feasibility of breeding many plants that will have relatively higher levels of nitrate reductase and hence lower levels of nitrates in the tissues. In summary, genetic control of nitrate uptake or reduction, or both, has been demonstrated in vegetables, including the feasibility of developing cultivars low in nitrate content.

### Neurotoxins and Lathyrogens

Lathyrism in man is characterized by muscular weakness and paralysis of the lower legs, disabling the victim and making it impossible for him to walk. This disease is associated with the consumption of seeds of Lathyrus species and is found in some parts of India and in the Mediterranean area. In India cultivation of this crop is banned, but it still persists as a weed in grain fields because of its high drought resistance. Consequently, it is eaten when the main crop fails. The problem of lathyrogens in foods has been carefully reviewed by Liener (18).

According to Swaminathan (30), lathyrism is caused by the neurotoxin  $\beta$ -N-oxalyl- $\alpha,\beta$  diaminopropionic acid (BOAA). He reported that natural populations of Lathyrus sativus ranged from 0.5 to 2.51 percent in the content of the toxic principle.

Swaminathan and his coworkers treated seeds of four commercial cultivars of L. sativus with X-rays and nitrogen mustard and selected individuals in the  $M_2$  generation for low content of BOAA. The mutants were true breeding for low content of BOAA. They were then subjected to a second cycle of treatment to isolate mutants even lower in content of BOAA. On the basis of data collected, the production of the neurotoxin was controlled by only a few genes. Apparently lines of Lathyrus could be developed that are essentially devoid of the toxic principle.

### Glycoalkaloids

Glycoalkaloids are a naturally occurring constituent of all potatoes as

well as other vegetables such as eggplant, peppers, and tomatoes. Alkaloids impart a bitter flavor to potato tissues and may be a factor in protecting plants from insects (28) and fungi (13). Some instances have been reported, primarily in Europe, where excessive amounts in potatoes have caused intestinal disorders and even death to humans and livestock (17).

In the normal potato tuber, most of the glycoalkaloid content is found near the skin and is consequently removed upon peeling, as shown in the following data:

TABLE 2.--Total glycoalkaloids (TGA) concentration in three varieties of potatoes

Potato variety	TGA concentration (mg./100 g. fresh wt.)		
	Peel	Flesh	Peel and flesh
Katahdin	81.0	2.3	10.1
Kennebec	76.0	1.5	9.7
Russet Burbank	69.0	1.2	8.0

The problem of glycoalkaloids in potatoes came to the forefront recently with the discovery of unusually high content in the potato breeding line B5141-6, formerly Lenape. This variety was released in 1967 by the U.S. Department of Agriculture and the Pennsylvania Agricultural Experiment Station because of its high solids content and adaptability for processing. Two years later scientists discovered that this variety consistently had a total glycoalkaloid content about twice as high as that of any other variety, as follows:

TABLE 3.--Total glycoalkaloids (TGA) in Katahdin and Lenape potatoes<sup>1/</sup>

Variety	TGA (mg./100 g.)	
	1968	1969
Katahdin	10.1	12.1
Lenape (B5141-6)	18.1	25.4

<sup>1/</sup> Source: unpublished data from Dr. S. L. Sinden, USDA.

As soon as it was realized that this variety had higher-than-normal glycoalkaloid content, the USDA and the Pennsylvania AES took immediate steps to remove this variety from the commercial trade (2).



Primarily as a result of the Lenape episode, research on glycoalkaloids in potatoes was expanded and new research initiated in both the United States and Canada, particularly on the nature of inheritance of glycoalkaloid content in potatoes. Sanford and Sinden (26) found significant differences in tuber glycoalkaloid (TGA) content among parents and among family means in a 2-year study of 10 tetraploid crosses. TGA contents of the parents ranged from 3.6 to 36 mg./100 g., with an average content of 10 mg./100 g. The average content in commercial varieties is about 8 mg./100 g.

Offspring variations within families were generally continuous, indicating polygenic inheritance. Sanford and Sinden concluded that TGA content was highly heritable and hence potato breeders could maintain low TGA contents in their breeding lines or even breed for a lower content of TGA.

Orgell and Hibbs (22, 23) studied in vitro cholinesterase inhibition by potato tissue extracts in many potato varieties and species. Extremely high human plasma cholinesterase inhibition was found in many species of wild potatoes, as well as a lower but wide range of readings among commercial cultivars. They believe that plasma cholinesterase inhibition probably reflected, at least in part, the presence of steroid alkaloids such as solanine.

Potatoes exposed to light turn green and become bitter as a result of increased levels of chlorophyll and glycoalkaloids. However, recently potato clones have been identified that do not turn green when exposed to light at intensities typically found in homes and stores, and in storage. Incompletely dominant multiple genes are involved in the inheritance of tuber greening (1). Studies have also been made to define the glycoalkaloid content in these greening-resistant clones (29) because light induces both glycoalkaloid and chlorophyll synthesis. Tubers of these clones synthesized less total glycoalkaloids and developed less chlorophyll than did tubers of clones that lacked resistance to greening. In similar manner, genetic variability for nongreening occurs in other vegetables, including carrots. The interrelationships of glycoalkaloid formation, cholinesterase inhibition, and nongreening require additional clarification before their significance can be ascertained, but we now have sufficient information on their genetic control to breed and select desirable types in this regard.

### Oxalates

Soluble oxalates when fed to 50-day-old rats completely immobilized body calcium (19). However, after a careful review of the literature, Fassett (12) questioned whether the reports on hazards to humans from the ingestion of oxalates in rhubarb leaves or in oxalate-containing vegetables seem warranted. On the other hand, Kingsbury (17) categorically states that ingestion of relatively small amounts of the rhubarb leaf blade is potentially lethal.

A study of 20 cultivars of spinach revealed highly significant differences in oxalic acid content, ranging from 8.66 to 10.52 percent on the dry-weight basis (10). These results suggest that spinach can be selected and bred for low oxalate content. However, the low oxalate lines of spinach are reported

flat in taste. Because of this and also because the hazards of oxalates are uncertain, low-oxalate spinach perhaps may not be a desirable breeding objective. In any case, we know of no serious attempt to develop low-oxalate cultivars.

#### Flatulence

Dry beans, lima beans, and related plants are often called the poor man's meat. They are a cheap source of protein for much of the underprivileged populations of the world, especially Latin America and Southeast Asia. However, many dry seeds of the legume family produce discomforting flatus when eaten by humans. Dry navy, kidney, and pinto beans produce especially high levels of flatulence (21). The ratio of flatus from bland test meal of 100 g. (dry weight) for a 3-hour period measured from 4 to 7 hours after ingestion as compared with flatus of different legumes is as follows:

TABLE 4.--Flatus from different legumes as ratio of flatus from bland test meal

Legume	Ratio
<u>Phaseolus vulgaris</u> L.	
California small white	11.1
Pinto	10.6
Kidney	11.4
<u>Phaseolus lunatus</u> L.	
Lima, Ventura	4.6
Lima, Fordhook	1.3
<u>Phaseolus aureus</u> Roxb.	
Mung	5.5
<u>Glycine max.</u> (L.) Merr.	
Soya, Lee or yellow	3.8
<u>Arachis hypogaea</u> L.	
Peanut	1.2
<u>Pisum sativum</u> L.	
Pea, dry	5.3
Pea, green	2.6
<u>Bland test meal</u>	1.0



Murphy also reported that there were significant differences in the flatulence factor in cultivars of dry lima beans, as follow:

TABLE 5.--Flatus produced by 3 cultivars of lima beans

Variety	Average flatus in 3 humans (total volume in cc. for 3-hr. peak period)
Fordhook - green mature	18
Fordhook - dry	39
Ventura - dry	227
Bland formula	23

The lima bean cultivar Fordhook (Phaseolus lunatus L.) was developed for use as a frozen product and is seldom eaten in the dry form, whereas the cultivar Ventura is primarily grown for the dry product. Therefore, the flatulence factor has been either lost in the development of the Fordhook cultivar or inadvertently added in the development of the Ventura.

An heirloom dry bean variety (Phaseolus vulgaris L.) has been kept in the Pike Family in New England for many years under the name "Jacobs Cattle" bean. Murphy (21) reported that this bean raised the formation of intestinal gas about four times that of the bland control as compared with the expected ten-fold response to the usual dry Phaseolus vulgaris group.

#### Low Digestible Starch

The quality of fresh sweet corn depends to a large extent on the sugar and polysaccharide content of the endosperm. The commercially available sweet corn cultivars derive their sugar and polysaccharide content through the genetic control of the recessive allele at the sugary-1 locus on chromosome 4. At optimum edible maturity, high quality sweet corn normally contains about 20 percent of its dry matter as sugar. Recently, Wann, Brown, and Hills (32) reported that total sugars could be increased markedly by using some of the endosperm mutants previously found by Creech (8); namely, ae (amylose extender on chromosome 5), du (dull on chromosome 10), and wx (waxy on chromosome 9). Their results are as follows:

TABLE 6.--Sugar content of sweet corn as affected by endosperm mutants

Corn selection	Total carbohydrates (percent)	Starch (percent)	Sugars (percent)
Golden Security (check)	75	32	22
M6212A ( <u>ae</u> <u>wx</u> )	73	35	35
M6210B ( <u>ae</u> <u>du</u> <u>wx</u> )	72	29	40

Thus, the new mutant genes were effective in raising the sugar levels in sweet corn to nearly double the amount found in the commercial sweet corn cultivar Golden Security. The production of total carbohydrates and starch meanwhile was slightly reduced, but essentially not affected insofar as the percentage of dry matter is concerned.

However, the starch content in the sugar corn lines with the mutant genes ae wx combination, when treated in vitro with pancreatic alpha amylase, was less digestible (25). Therefore, from these rather preliminary tests we might state that the combination of endosperm mutant genes ae wx had the pleiotropic effect. It appeared to double the sugars, while at the same time changing the starch structure to a form poorly digestible by the enzyme that normally digests starch in the small intestine. However, these results are preliminary and to my knowledge have not been verified by in vitro tests.

One might ask whether low digestible starch is an undesirable characteristic in the American diet, and human nutritionists probably would pause for a great deal of thought on the matter.

### Bitter Principle in Cucurbits

Vegetables of the Cucurbitaceae family possess genes that control bitterness factors in their fruits, particularly cucumbers, squashes, and watermelons. Twelve bitter principles, known as cucurbitacins, have been found (24). Plant breeders have no difficulty in selecting and breeding nonbitter fruits. However, these substances are of interest because they have been reported to be carcinostatic (14) as well as attractants for spotted cucumber beetles (Diabrotica undecimpunctata howardi Barber) (7).

Bitterness is apparently genetically controlled in watermelon by a complex system of a single recessive suppressor gene su bi that is active in the presence of the dominant gene for plant bitterness, Bi, along with a modifier gene Mo bi that modifies the amount of bitterness (6).



## Other Objectionable Qualities Possibly Controllable Genetically

Other undesirable compounds or unattractive characteristics are as follows:

<u>Name of compound or characteristic</u>	<u>Mode of inheritance</u>	<u>Authority</u>
Pubescence in peaches	Single dominant factor between nectarines and peaches. Heavy pubescence is dominant over light pubescence.	Blake and Connors (4)
Pit astringency in apricots	Probably multifactorial.	Cullinan (9)
Antimetabolites in beans	Not known, but there are varietal differences.	Unpublished data
Favism	Inheritance of the factor in <u>Vicia faba</u> L. is unknown. In susceptible humans the suscepti- bility to favism is apparently sex-linked factor with incom- plete dominance.	Zinkham, Lenhard, and Childs (34)

### Summary

Almost all available papers on genetic research to remove undesirable compounds from vegetables and fruits have been published since 1970. Limited knowledge is available on the mode of transmission of genes in vegetables governing such traits as nitrate accumulation and formation of neurotoxins and glycoalkaloids. Likewise, unattractive characteristics of vegetables and fruits such as flatulence, bitter taste, excessive pubescence, and unattractive color change are apparently genetically controlled, but the gene action governing these conditions is not well understood. This paper has reviewed essentially all known research to date on genetic engineering to remove these undesirable compounds and unattractive characteristics.

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## EFFECTS OF GENETICS AND DIET ON MEAT COMPOSITION

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In considering meat composition it is logical to start with a consideration of animal composition. Animal bodies are for the most part composed of water, fat, protein, and ash. Among these the chief variable in body and carcass composition is fat, and variations in the concentration of other components primarily reflect the changes in the amounts of fat.

Detailed evaluation was made of the body composition of 256 cattle of widely different ages, including approximately equal numbers of males and females and approximately equal numbers of cattle of beef and dairy breeding types. The study showed rigid, predictable relationships between body components (13). In this study the correlation coefficients between body water and fat were high, -0.985 to -0.989. This relationship was not noticeably influenced by breed type or sex. Animal scientists can predict the body composition from estimates of total body water (13).

Wide ranges exist in the concentration of ether-extracted fat in the bodies of farm animals. Reid (10) reported pigs to range from 1.1 to 61.5 percent, sheep from 4.9 to 46.6 percent and cattle from 1.8 to 44.6 percent. The extent of the influence of body weight, type, and sex on body and carcass composition varies with species. In a detailed study of the chemical composition of 714 pigs representing nine breeds, a variety of dietary treatments, and a great range in body size, it has been noted that intact female pigs contain more fat than male castrates from birth to the time they reach 70 kg. Above 70 kg., barrows are fatter than gilts (11).

The extent of the influence of extreme variation in the body type of swine on composition was studied by Mitchell and Hamilton (9). When the reported weights of the body components were adjusted by covariance analysis to a common empty-body weight of 100 kg., water ranged from 45.2 to 47.1 percent, fat from 40.9 to 38.1 percent, protein from 12.9 to 13.1 percent, and ash from 2.3 to 2.4 percent in the bodies of the chuffy, intermediate, and rangy body types (12). Mitchell and Hamilton (9) stated that carcass analysis revealed only inappreciable differences between types in spite of large differences in the apparent market finish. Recently Hertzner and Miller (7) described high and low backfat thickness strains of swine in which the high-strain Durocs had a



49 percent divergence in backfat thickness from the low strain, and high-strain Yorkshires a 20 percent divergence. Doornenbal *et al.* (4) reported that, as an index of total carcass fatness or of chemically determined fat, backfat thickness is an inadequate indicator. Doornenbal's study showed that only 13 to 38 percent of the variance in the chemically determined fat of the half carcass is associated with the variability in the probe-measured backfat thickness.

The inescapable influence of body size on body and meat composition has been clearly demonstrated. Burton and Reid (3) imposed two decidedly different supermaintenance levels of energy input on sheep so that they attained given body weights at markedly different ages. They observed that almost all of the variation in body composition was associated with body weight and very little was ascribable to age. George *et al.* (6) demonstrated that as lambs grew and fattened from 33 to 60 kg., although they became fatter, the extra fat trim on the carcasses almost completely accounted for the greater dressing percent of the heavier lambs. The percentage of the original live lamb included in four trimmed retail cuts (leg, loin, rack, and shoulder) was not greatly affected, however, by body weight. The wholesale ribs from the heavier lambs did have a greater percentage of ether-extractable lipids and a lower percentage of water and protein. Cooking tended to diminish the differences. The illustrations of composition changes cited here for sheep are in general agreement with similar changes that accompany increases in body weight in swine and cattle.

The influence of differential rates of growth on carcass composition of various body parts has been reported for the pig in extreme detail by McMeekan (8) and growth curves have been developed to represent, respectively, bone growth, muscle growth, and fat deposition. Even though these differential growth processes occur earlier where the plane of nutrition is higher, the practical application of these principles within the American system of swine production seems doubtful. Berg and Butterfield (2), after much study by muscle dissection of cattle carcasses, have concluded that major differential growth within the bovine musculature takes place soon after birth and would play a minor role in affecting the composition of animals of normal slaughter weights.

The offspring of larger cattle breeds are relatively less mature at a fixed slaughter weight, for example 1000 pounds, than those from smaller breeds. Since less maturity is associated with less fat, the use of beef sires from large breeds or strains offers a method for producing less fat beef carcasses at an established slaughter weight. British studies have illustrated the influence of potential body size in a carcass comparison of Fresian and Hereford cattle (1). From 12 months of age, Fresians had heavier carcasses with more muscle than the Herefords in the study. When the fattening process began, the Hereford steers fattened at lighter weights than Fresians. Although the differences thus recorded were reported for specific breeds, the suggestion is strong that these expressions of differences in growth and fattening were expressions of differences between larger and smaller bovine types. More basic information on bovine growth and fattening, expressed in both chemical and physical units, will help complete the



understanding of the influence of type and nutrition on bovine composition.

Beef sires of large scale from previously little-known breeds have attracted unusual attention among beef breeders. Their offspring can be expected to grow rapidly and be less fat at a fixed slaughter weight. Adams has compared rates of feedlot gain between cattle sired by bulls of conventional breeds and those sired by some of the unusual or "exotic" breeds. Cattle from Simmental, Maine-Anjou, Brown Swiss, Limmousin, and Charolais sires had greater average daily gains than cattle sired by Angus and Hereford bulls. Fat composition calculated from carcass density indicated that cattle sired by Angus and Hereford bulls had carcasses that were fatter than cattle sired by Brown Swiss, Simmental, Limmousin, Maine-Anjou and Charolais sires. Carcass from cattle sired by Angus, Hereford, and Brown Swiss were USDA Choice in grade and those sired by Charolais, Simmental, Limmousin, and Maine-Anjou were USDA top Good at slaughter. (Personal communication from N. J. Adams, of Santa Rosa, Calif., who, with others at Texas A&M University, has submitted this research for publication in the Journal of Animal Science.) The present preference within the meat trade for USDA Choice beef over Good grade is pronounced and is not likely to quickly change. The meat tradesmen have tended to anticipate negative consumer response to beef that graded lower than Choice.

Much has been reported on the influence of nutritive protein levels of meat composition. With pigs, the extent to which the chemical composition of the whole body or carcass can be changed through changes in quantity or quality of protein remains unsettled (10). The effects have not been clearly defined because in general the criteria of leanness or fatness has been subjective, and fat infiltration in lean tissue has influenced lean yield data. In addition, physically separated pork fat may contain as much as 90 percent and as little as 30 percent ether extract. As cited earlier, backfat thickness is not infallible as an indicator of fatness. Studies by Wallace (14) indicate that a diet for pigs of 17 percent protein, followed by 15 percent protein, is advantageous over a 13-11 percent protein sequence. Both barrows and gilts gained significantly faster and more efficiently and yielded carcasses with less backfat, larger loin eye areas, and greater percentages of lean cuts when they were fed at the higher protein levels. Visual appraisal of loin eye marbling revealed much more intramuscular fat deposition in pigs fed the 13-11 percent sequence of protein. The differences reported were significant, but modest in amount.

Epley et al. (5) reported a beef feeding experiment with 42 Herefords on isocaloric diets to study the effects of the ratio of digestible protein to digestible energy on carcass composition. The DP:DE ratios used were 10:1, 22.1, and 27.6. There was essentially no effect of nutritional treatments on carcass fat thickness, longissimus muscle area, conformation score, marbling score, quality grade, yield grade, weight of trimmed, boneless cuts, weight of fat trim, tissue ether extract, or protein.

Research designed to give both carcass data and body composition data on individual animals is involved and expensive. However, such studies will give much more precise answers to the influence of inheritance and diet on meat composition.



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## EFFECT OF COW'S DIET ON MILK COMPOSITION

(Condensation)

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Milk is a highly nutritious food that is composed of a complex array of chemical entities; an array that provides all of the nutrients required by the rapidly developing, nutritionally demanding neonate. It is not surprising that alterations in the cow's diet can change the composition of milk because all of the wide variety of chemical entities found in milk are ultimately derived or synthesized from constituents of the cow's diet, and cows are fed a great diversity of feedstuffs.

The most compelling evidence that feed can affect the composition of milk is obtained when off-flavors are transferred from feed to milk, such as occurs when cows graze wild onions in the spring. Modern dairy processing has alleviated that problem. More important are changes that can occur in the major nutrients of milk--the protein, fat and carbohydrate. While our interest here is in dietary effects on milk composition, it is well to bear in mind that genetic, environmental, physiological, and management factors also influence milk composition; these effects are not always rigidly controlled when studying dietary effects.

The most variable component of milk and the most studied in relation to diet is the butterfat. Interest in the effect of diet on the butterfat content of milk is understandable because half of the caloric value of milk is associated with the fat, and pricing systems for milk have been based on the fat test. More recently the relation of the human diet, and specifically the fat in the human diet, to heart disease has added a new impetus to studies of milk fat.

Dietary effects on the fat content of milk can indeed be dramatic. Cows producing milk containing 3.5 to 4.0 percent butterfat when fed "normal" diets have been observed to produce milk containing less than 1 percent butterfat when fed high-energy, low-fiber diets. Other dietary regimes that have caused decreased milk fat secretion involve the grazing of certain kinds of pasture grass, spring oats and pearl millet, and the feeding of certain unsaturated

fatty acids. Short-term increases in butterfat can be obtained by including supplemental fat in dairy cow rations.

The protein in milk is now recognized as the most important nutritional constituent of milk, and some milk markets are giving it appropriate credit by tying milk prices paid to dairymen to the protein content of the milk. The variation in protein concentration of milk is not nearly so large as that found in the fat content; nevertheless, variations do occur in response to changes in the nutritional status of the cow. If protein content of all the milk produced annually in the United States was changed from 3.2 percent to 3.4 percent, the total production of high-quality protein would increase by 55,000 tons.

Other minor components of milk, including many of the important vitamins and minerals, may also vary in amount in response to dietary changes. Our affluent position with respect to food supplies in the United States has not provided the economic pressures to explore such possibilities. Factors affecting the content of minor nutrients in milk have not been adequately investigated, nor have dairymen made the necessary inputs to produce milk with optimal amounts of these constituents. As the relationship between population and available food supply becomes more critical, attention will be brought to bear on the total nutritional value of milk.



## INCREASING THE UNSATURATION IN MILK FAT AND IN CARCASS FAT OF MEAT ANIMALS

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The effect on our health of saturated and unsaturated fats in our diet is not clear. All of us are aware, however, of reports from medical sources recommending that we lower our overall consumption of fats and increase the relative amounts of polyunsaturated fats (e.g., linoleic acid) in our diet. The reason, of course, for such recommendations relate to correlations reported between the high intake of saturated fats and the incidence of heart diseases.

Milk and ruminant meat fats are relatively high in saturated fat and contain very little linoleic acid, an 18-carbon acid containing two double bonds (C 18:2). An obvious method of increasing the polyunsaturation in milk is to add a vegetable oil, or replace a part of the milk fat with such an oil, followed by pasteurization and homogenization. This procedure, however, would have little or no application for modifying meat fat.

This discussion reviews research underway in the Agricultural Research Service, U.S. Department of Agriculture, on a technique to increase the polyunsaturation in milk and meat fat by feeding ruminant animals a supplement of encapsulated, or "protected," vegetable oil. This work is a cooperative project being carried out by the Animal Physiology and Genetics Institute, Beltsville, Md., and the Dairy Products Laboratory, Washington, D. C. The studies on veal fat reported herein were done by members of the Meat Laboratory, Eastern Regional Research Laboratory, Wyndmoor, Pa. The technique was first reported by Scott and associates (5, 6) in Australia. Results discussed herein were reported earlier this year at the annual meeting of the American Dairy Science Association (2, 8) and have been submitted for publication (1, 3, 7).

### Preparation of Feed Supplement

The "protected" vegetable oil (safflower, or other suitable oil) was

prepared by homogenizing the oil with a solution of sodium caseinate (about 10 percent casein, 70° C.), treating with 37 percent formaldehyde (6 to 10 percent by weight of protein), and spray drying. The oil and formaldehyde may be metered into a continuous flow line of caseinate before homogenizing; alternatively, for small lots, the oil may be added to the caseinate solution in a vat followed by homogenization. In the latter case, formalin is added slowly to the homogenized blend with thorough stirring.

Formaldehyde reacts with the protein (casein) to form a coating around the fat globules. This coating is stable at the pH of the rumen and protects the encapsulated fat from microbial hydrogenation. The formaldehyde-protein matrix is hydrolyzed in the more acidic conditions of the abomasum, allowing the polyunsaturated fatty acids to be absorbed.

Among the objectives of various programs of feeding "protected" fat are to determine: the effect of the amount of the protected supplement given dairy cows on the amount of C 18:2 in the milk fat, the quality of milk and milk products resulting from feeding "protected" fat, and the effects of feeding veal calves rations (milk and dry feed) that contain high levels of C 18:2 on the C 18:2 content and oxidative stability of rendered fat from these animals.

#### Effect of Amount of Protected

##### Dietary C 18:2 on Amount in Milk Fat

Two Holstein cows were fed a standard ration of hay and concentrate, supplemented with varying amounts of protected safflower oil [SOC-F] for six consecutive weeks. Each week the amount of SOC-F fed daily to each cow was twice the daily feeding level for the preceding week. The amounts were: 0, 200, 400, 800, 1600 and 3200 grams per day. The SOC-F supplement contained about 65 percent safflower oil, which assayed 75 percent C 18:2. Because of feed refusals, the average daily intake of SOC-F for the two cows was 762 and 2278 grams per day during the fourth and sixth weeks, respectively, instead of the planned amounts of 800 and 3200 grams per day.

Figure 1 shows weekly changes in some of the major fatty acids of the milk fat. The C 18:2 content increased from less than 3 percent of total fatty acids in normal milk to 30 percent in milk from the cow fed at the highest SOC-F level. This is indeed very striking. A smaller increase also occurred in the C 18:1 acid, even though safflower oil contains less of this acid than normal milk fat. This result may be due to partial hydrogenation of C 18:2. A marked compensatory decrease occurred in the C 16:0 acid. Also, a smaller, but significant, decrease was observed for C 14:0.



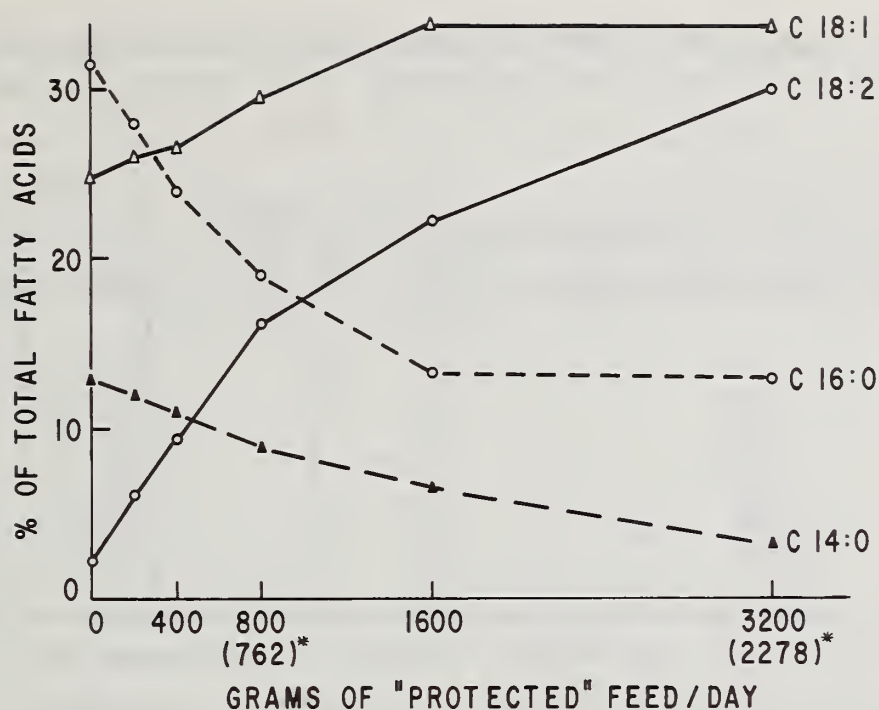


Figure 1.--Effect of amount of "protected" feed (SOC-F) in cow's ration on changes in fatty acids in milk fat.

#### Effect of Feeding the SOC-F Supplement on the Flavor of Milk

Pooled samples of milk, taken at weekly intervals, from the two cows on the feeding regimen described above were also evaluated for flavor. For this purpose, the milk was pasteurized and homogenized, and scored by a 10-man panel after 1, 8, and 15 days of refrigerated storage. Results are given in table 1. The flavor score decreased with increasing amounts of C 18:2, and decreased further during storage for most samples. Off-flavor criticisms were predominantly of an oxidized type.

A study made to determine whether oxidation occurred in the SOC-F supplement prior to feeding, or whether it developed in the freshly drawn milk, showed that the intensity of oxidized flavor was negligible in raw milk immediately after milking, but increased markedly after 24 hours of refrigerated storage (figure 2). Samples which were pasteurized and homogenized immediately after milking (within 2 hours) were slightly more oxidized than raw milks, and exhibited a further gradual increase during storage. (The intensity ratings are taste panel averages based on the following scale: 0 = none, 1 = questionable, 2 = slight, 3 = definite, 4 = strong. Milk for this study was the pooled collection from 4 cows, and contained about 12 percent of C 18:2 in the milk fat.)

The lack of any significant oxidized flavor immediately after milking posed the obvious question: would the addition of an antioxidant to the fresh milk be beneficial? This problem was studied by adding alpha tocopherol

TABLE 1.--Flavor scores of milks obtained from cows fed stepwise increases in the level of "protected" safflower oil<sup>1/</sup> during succeeding weeks

Feeding program		Milk sample No. <sup>2/</sup>	Flavor score <sup>3/</sup>		
Week	"Protected" safflower oil fed (grams/day/cow)		1 Day	8 Days	15 Days
1st	0	1	37.2	35.7	35.8
2nd	200	2	36.3	33.8	35.2
3rd	400	3	35.3	36.5	34.7
4th	762	4	35.4	33.8	32.3
5th	1600	5	34.5	34.1	32.7
6th	2278	6	32.0	31.5	32.0

<sup>1/</sup>"Protected" supplement contained 65 percent safflower oil.

<sup>2/</sup>Pooled milk from 2 cows, collected at mid-week of each interval, pasteurized and homogenized.

<sup>3/</sup>Scores are average of a 10-judge panel. Scores ranged from 31 to 40. Score of 35 considered acceptable.

(50 micrograms per gram of fat, emulsified in Triton X-100) to the freshly drawn milk. Milk available for this study contained only 8 percent C 18:2. Nevertheless, a definite oxidized flavor developed in the raw milk without antioxidant within 3 days, whereas this off-flavor was insignificant in samples to which tocopherol was added. This problem will be further studied to determine the effectiveness of antioxidants in milks containing 20 to 25 percent C 18:2, and to determine whether antioxidants impart a characteristic off-flavor of their own.

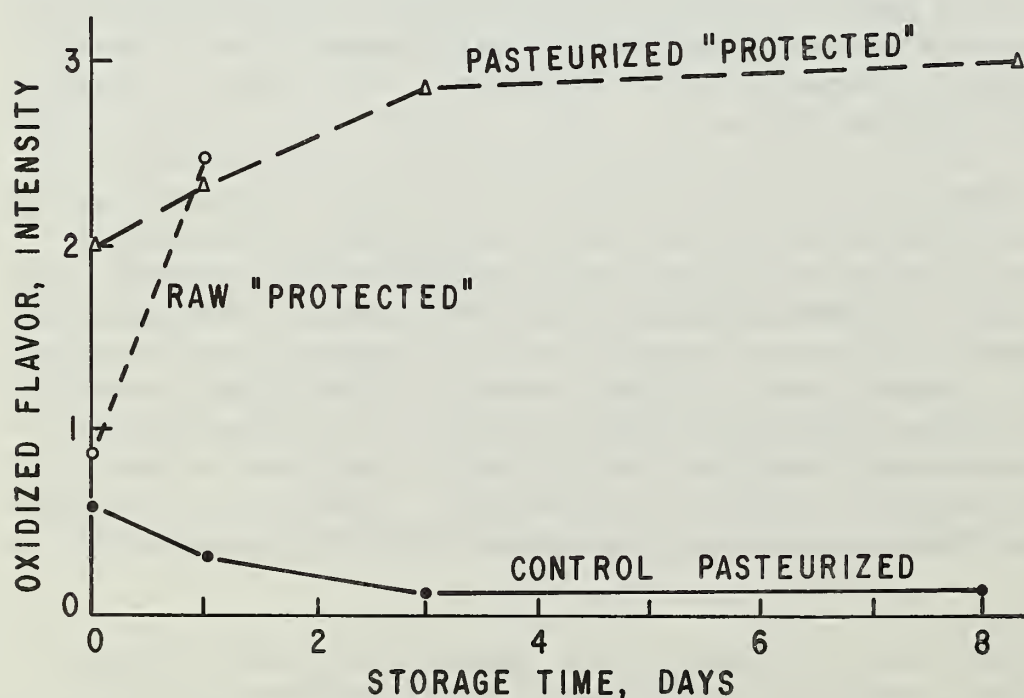


Figure 2.--The effect of feeding "protected" safflower oil to cows on the development of oxidized flavor in the milk.



## Physical and Chemical Properties of

### Milk Fat with High Levels of C 18:2

Milk fat from cows fed SOC-F showed the changes in physical and chemical properties expected of a highly unsaturated fat. For example, the iodine value shifted from 32 for normal milk fat containing 2.1 percent C 18:2 to 79 for milk fat containing 30 percent C 18:2. Melting profiles, determined by differential thermal analysis, showed a shift in the inception melting point from  $-21^{\circ}$  C. for the normal fat to  $-43^{\circ}$  C. for the highly unsaturated fat. Cream containing 30 percent of C 18:2 in the fat required a longer aging time and lower aging temperature for churning into butter than normal cream. The butter was soft and somewhat sticky, but was readily spreadable at refrigerator temperatures, and exhibited a distinctly lighter color than normal butter. Creams for whipping also required changes in whipping conditions and produced lower overruns.

Gas chromatographic analysis of the triglyceride content of polyunsaturated milk fat (30 percent C 18:2) showed an expected high proportion of triglycerides containing fatty acids with a total of 54 carbon atoms, in contrast to the proportion of  $C_{54}$  triglycerides in normal milk fat (4). However the  $C_{40}$  triglyceride content remained the same (about 12 percent of all triglycerides from  $C_{28}$  to  $C_{54}$ ). Analysis of the fatty acid composition of the  $C_{40}$  triglyceride component showed a much higher proportion of C 18 acids (including 18:2) in polyunsaturated fat than in normal fat. The difference in fatty acid distribution patterns was such as to indicate that the dietary "protected" safflower oil was not transferred to milk in its original triglyceride state, but that hydrolysis and resynthesis occurred before incorporation into the milk.

### Meat Fat with Increased Levels of C 18:2

The polyunsaturation in meat fat of ruminant animals can also be greatly increased by feeding "protected" vegetable oils. In one experiment, eight 4-day old bull calves were divided into two groups. Four of the calves were fed milk containing 14 percent C 18:2 in the milk fat for 10 weeks; the other four were fed normal milk (3 percent C 18:2 in the milk fat). Both milks were supplemented daily with d-alpha tocopherol. After 10 weeks, two calves from each milk group were transferred to an 8-week dry-feed regimen which included the spray-dried formaldehyde-treated, caseinate-safflower oil supplement (SOC-F). The other two calves from each milk group were fed a dry ration containing caseinate-safflower oil (SOC, not formaldehyde treated). After 18 weeks the calves were slaughtered. Analyses for fatty acid composition of rendered round fat are given in table 2. The tocopherol contents and oxidative stability of the fats are given in table 3. The results show a 4-fold increase in the C 18:2 content of the fat of the two calves fed milk high in C 18:2 followed by "protected" dry feed, compared to the C 18:2 content of the calves given normal milk and "unprotected" dry feed. The proportion of

TABLE 2.--The fatty acid composition of the veal depot fat from bull calves fed milk and then a dry feed supplement with low and high levels of polyunsaturated fat

Feeding regimen of calves		Composition of major fatty acids in veal depot fat (weight percent) <sup>1/</sup>				
Milk (1st 10 weeks)	Dry feed supplement (11th to 18th week)	14:0	16:0	18:0	18:1	18:2
Normal (3 percent C 18:2)	Safflower oil-caseinate blend					
	Not treated	6.7	31.5	16.4	37.2	3.1
	Formaldehyde treated	5.8	28.5	18.7	34.5	8.2
Polyunsaturated (14 percent C 18:2)	Safflower oil-caseinate blend					
	Not treated	4.3	22.4	17.4	41.0	10.2
	Formaldehyde treated	5.1	24.6	17.1	36.3	12.6

<sup>1/</sup>Average of fat from 2 calves.

TABLE 3.--The effect of C 18:2 fatty acid and tocopherol content on the stability of veal fat

Feeding regimen of calves		Rendered round fat <sup>1/</sup>		
Milk (1st 10 weeks)	Dry feed supplement (11th to 18th week)	Tocopherol ( $\mu$ g./g. of fat)	C 18:2 fatty acid (percent)	Induction period (days)
Normal (3 percent C 18:2)	Safflower oil-caseinate blend			
	Not treated	30.1	3.1	45
	Formaldehyde treated	33.4	8.2	27
Polyunsaturated (14 percent C 18:2)	Safflower oil-caseinate blend			
	Not treated	25.6	10.2	19
	Formaldehyde treated	57.0	12.6	23
Normal feeding regimen (commercial veal fat)		8.5	5.5	15

<sup>1/</sup>Average of fat from 2 calves.



C 16:0 decreased with an increase in C 18:2 (a compensatory effect).

The tocopherol levels in the round fat of the experimental calves were from 3 to 7 times that of a commercial veal sample (table 3). An explanation for the variation in tocopherol content of the fat from the experimental animals (from 30 to 57 micrograms per gram of fat) is not obvious (the milk of all calves was supplemented with tocopherol). However, fat samples from the experimental calves taken as a group were decidedly higher than commercial veal fat both in tocopherol content and in days required for induction period. In evaluating fats, a shorter induction period is considered to denote lower stability against oxidation.

The technique of increasing the polyunsaturation in milk and meat fat of ruminant animals is in the experimental stages. Before its feasibility can be established, its economic aspects must be studied, more consistent results must be achieved, an appropriate means must be found to overcome the tendency of highly unsaturated fats to undergo oxidative changes, and the effects of the special feed on the health of animals must be explored. Further studies on the role of fats in the human diet and the extent to which changes should be made will also be a factor in decisions concerned with milk and meat fat modifications.

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## EFFECTS OF DIET AND OTHER FACTORS ON THE NUTRIENT COMPOSITION OF POULTRY PRODUCTS

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Meat and eggs from chickens, ducks, and turkeys are important human foods. Although serving primarily as sources of protein, they also contribute substantial quantities of fat, minerals, and vitamins. All are important nutrients, but recent research and public interest have focused largely on their fat, fatty acids, and cholesterol content. There is a steadily expanding pool of knowledge on the effects of diet, management, and genetics on these compounds but unfortunately progress has been slowed by confusing and contradictory results. It is the objective of this talk to comment briefly on some aspects of this broad and very complex subject.

### Composition of the Egg

Differences in egg composition due to the genetic background of the bird were first reported by Scrimshaw et al. (21), and confirmed by Howes and Hutt (14), with the observation that Leghorns secrete 60 percent more thiamine into their eggs than do heavy breeds. Arroyave et al. (1) found significant differences among the eggs of five breeds in the total content of nitrogen, phosphorus, ash, carotene, and vitamin A. Most of the differences, however, were relatively small and did not appear to have practical implications. Taylor (25), after an extensive review of the literature, was led to remark that of all the egg structures, the yolk is least influenced by hereditary factors. Despite the apparent difficulty of achieving yolk modification via genetics, interest in this approach has continued. It was reported by Edwards et al. (6), and confirmed by Collins et al. (3), that significant differences exist between strains or breeds of chickens in egg yolk cholesterol level. Bartov et al. (2) noted that birds laying at a higher rate secrete more total cholesterol, but less cholesterol per egg yolk than do birds laying at a lesser rate. Earlier, however, Collins et al. (3) had noted that the variation between strains was at least partially independent of rate of lay. Harris and Wilcox (12) concluded that although differences between birds or strains did exist,

egg yolk cholesterol was not a highly heritable trait. In full agreement, Estep et al. (7) estimated the heritability of serum cholesterol in birds one year of age to be only 0.26, based on the regression of offspring on sire.

Shaklee (24) reviewed 51 cholesterol studies published during the previous 5 years, and noted many conflicting results. He concluded, however, that increased dietary cholesterol (usually massive doses) resulted in increased serum and egg yolk cholesterol and atherosclerosis in chickens. Exercise or an increased heart rate induced by a pacer was accompanied by a decreased atherosclerosis score. Because of the many conflicting results, Bartov et al. (2) attempted to define more accurately the experimental parameters that might be used in cholesterol studies. They confirmed previous work, reporting a highly significant variation in yolk cholesterol between hens within a flock, but also noted a highly consistent yolk cholesterol level for the same hen. They also noted a marked fluctuation in plasma cholesterol in the same hen as well as between hens in the same flock. Over a six-day period, plasma cholesterol measured in the same hen showed variations ranging from 20 to nearly 75 percent with an average of 49.7 percent. During this same period, yolk cholesterol varied only from 3.0 to 8.6 percent with an average of 5.7 percent. They concluded that plasma cholesterol was essentially useless in predicting egg yolk cholesterol content, and that it would be of highly questionable value in measuring treatment results in small groups of hens. As an alternative, they proposed that each hen should serve as her own control, and that treatment effects should be measured relative to the pre-treatment period. In light of these results, it is not surprising that many conflicting data have been reported during recent years, and it is suggested that many of the studies will need to be redone.

Of particular interest are the reports by Griminger and Fisher (11) and Fisher and Griminger (8, 9) that pectin and some grain fractions reduce blood cholesterol. Ground whole oats, wheat, and barley were effective, while corn had no effect. Oat hulls were most active, while oat starch and oat oil were without effect. Pectin also reduced atherosclerosis in long-range tests with male chickens.

It is well established that the fatty acid composition of egg yolk fat reflects the composition of the fatty acids in the diet. Hill and Peterson (13) noted that eggs from hens fed a typical practical diet will contain from 15 to 20 percent linoleic acid. Levels ranging from a low of 6 to more than 40 percent have been reported. Wheeler et al. (26) reported that feeding from 10 to 30 percent of a highly unsaturated fat such as corn oil or safflower oil may increase yolk linoleic acid to approximately 40 percent of the total yolk fat. My own calculations based on their data indicate that the efficiency of transfer of feed linoleic acid to the yolk was approximately 27 percent regardless of the level or source fed. Because high levels of dietary oils are physically undesirable and because the efficiency of transfer of linoleic acid is relatively low, it would appear impractical to attempt large-scale product modification via this route.



## Composition of the Tissue

For all practical purposes, the composition of tissue protein defies change. The quantity of protein produced can be increased by diet, management, and genetic selection, but the amino acid composition remains stable. This is a substantial advantage to the poultry industry, since it is possible to assure the consumer that protein quality has not been altered by increased production.

Heritable differences in the amino acid utilization have been reported, but no information is available on the effect of these differences on the composition of the products produced. McDonald (15, 16, 17) reported a breed difference in methionine utilization due to a sex-linked, dominant gene that affected ability to convert methionine to cystine. Singsen *et al.* (22) and Godfrey (10) observed wide differences in the requirements of individual birds for lysine. Godfrey's attempt to select for differential lysine utilization in 10 generations of Japanese quail yielded poor results. Heritability of this trait was estimated at only 0.31. Nesheim *et al.* (19) summarized several years of study on the biochemical basis for strain differences in arginine and lysine utilization in the chick. These differences are known to alter the dietary requirement for these amino acids, but their effect on the composition of the product produced has not been elucidated.

The effects of diet on body composition have been reviewed by Hill and Peterson (13). The principal factors involved are: (1) total energy intake, (2) protein-to-energy ratio, and (3) quantity and composition of dietary fat. These effects are well illustrated by the studies of Donaldson *et al.* (5), Summers (23), and Marian and Woodruff (18) with chickens; Dean (4) with ducks; and Salmon and O'Neill (21) with turkeys. With all birds, carcass fat increased as dietary energy intake increased. A wide calorie-to-protein ratio resulted in increased fat deposition, reflecting the relative increase in energy intake. In sharp contrast, changes in dietary protein had little effect on carcass fat when all rations were maintained isocaloric. In all cases, total tissue fat and moisture were inversely related, while the quantity of moisture-free/fat-free solids was relatively stable.

The problem of excess carcass fat and consequent loss to both the processor and consumer has also received attention. Excess fat may be trimmed away at the dressing plant and, in addition, present day calorie-conscious consumers will remove and discard all possible fat at the point of consumption. Both practices represent a loss of useful food energy as well as a loss of money paid for a product. An extreme situation is illustrated by the data of Dean (4). Ducks fed similar quantities of protein, minerals, and vitamins, but varied energy allowance ranged from 4.22 to 6.26 pounds body weight at 7 weeks of age. Analytical data revealed that the 2.04 pounds of additional weight contained only 0.18 pound of moisture-free, fat-free solids, including bone. In light of the practice of discarding fat, the consumer would receive very little usable material in the additional 2.04 pounds purchased.

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